# National Antimicrobial Resistance Monitoring System (NARMS) Internal Review and Recommendations

U.S. Food and Drug Administration
United States Department of Agriculture
Centers for Disease Control and Prevention

Submitted by the
NARMS Internal Review Committee
to the FDA Science Board

March 31, 2006

# Table of Contents

Background……...…….....3

Sampling……....6

Epidemiological and Microbiological Research …….……17

Harmonization of Data Reporting/Recording …….22

Coordination with International Surveillance …….25

Questions for the Science Board……. ……28

Appendix I-August 2003 CDC's Expert Review of Human Component…….....29

Appendix II- June 2005 Expert Review - 3 arms (FDA, USDA, CDC)……..……45

Appendix III- NARMS Internal Review Committee Members……...52

Appendix IV- NARMS Research References…….....53

Appendix V- Examples of Tables and Figures for NARMS Integrated Report……..57

## BACKGROUND

In food animals, antimicrobials are used for the control, prevention and treatment of infectious bacterial diseases, as well as for enhancing growth and feed efficiency purposes. An undesired consequence of antimicrobial use in animals is the potential development of antimicrobial-resistant zoonotic foodborne pathogens, and their subsequent transmission to humans via foods.

Recognizing this potential health hazard, the World Health Organization (WHO), the Food and Agriculture Organization (FAO) and the World Organization for Animal Health (OIE) recommend that each country implement a monitoring program on both the usage of antimicrobials in animals as well as on the occurrence of antimicrobial resistance in bacteria from animals and from food of animal origin (http://whqlibdoc.who.int/hq/2004/WHO_CDS_CPE_ZFK_2004.7.pdf).

WHO, FAO and OIE recognize that data obtained by such monitoring may be used to:

- document the usage of antimicrobials and the occurrence of resistance and to identify epidemiological trends;
- compare the usage of antimicrobials and the occurrence of resistance between countries or regions and between time periods;
- aid interpretation of patterns and trends regarding antimicrobial resistance and residues;
- develop risk assessment;
- evaluate the effectiveness of any control measures implemented;
- identify focused and targeted research;
- develop policies for the containment of antimicrobial resistance.

As part of the overall Center for Veterinary Medicine (CVM) strategy to assess relationships between antimicrobial use in agriculture and subsequent human health consequences, the National Antimicrobial Resistance Monitoring System (NARMS) program was developed in 1996 to monitor changes in susceptibility of select bacteria to antimicrobial agents of human and veterinary importance. NARMS is a collaboration between 3 federal agencies including FDA's Center for Veterinary Medicine (CVM), the Centers for Disease Control and Prevention (CDC) and the U.S. Department of Agriculture (USDA). NARMS also collaborates with antimicrobial resistance monitoring systems in other countries, including Canada, Denmark, France, the Netherlands, Norway, Sweden and Mexico so that information can be shared on the global dissemination of antimicrobial resistant foodborne pathogens.

The NARMS program monitors antimicrobial susceptibility/resistance among two categories of enteric bacteria recovered from food animals, humans, and retail meats: zoonotic bacterial pathogens (*Salmonella* and *Campylobacter*) and commensal bacteria (*E. coli* and *Enterococcus*). All three arms also characterize *Salmonella* (and *Campylobacter*) through use of Pulse-Field Gel Electrophoresis (PFGE) in an effort to determine genetic-relatedness between isolates. Epidemiological and microbiological

research studies are conducted within each agency or between agencies on isolates of special interest such as those of a particular serotype or expressing a particular resistance pattern. Currently each NARMS agency prepares a comprehensive annual report with a large quantity of data available on each agencies website. Data and targeted research studies are reported at scientific meetings and published in peer reviewed scientific journals.

**NARMS Goals**

- **generate descriptive data on the extent and temporal trends of antimicrobial susceptibility in enteric organisms from the human and animal populations**
- **provide information to veterinarians, physicians, and public health authorities on unusual or high levels of bacterial drug resistance so that timely action can be taken to protect public health**
- **design follow-up epidemiology and research studies to better understand the emergence and transfer of antimicrobial drug resistance**
- **prolong the life span of approved drugs by promoting the prudent use of antimicrobials**

## Previous NARMS Reviews

### CDC NARMS Review

In August of 2003 (Aug 12-13, 2003), CDC convened an external review meeting to review the CDC arm of NARMS. Panelists with expertise in food safety, microbiology, surveillance and epidemiology participated in reviewing and evaluating the CDC NARMS component. The panelists were asked to respond to specific questions that were believed critical to the future success of CDC's NARMS programs. The external review report, along with CDC's responses to the panelist's comments, are provided in their entirety in Appendix I of this document.

### NARMS Expert Review

In June 2005 (June 23-24, 2005), CVM convened a meeting requesting specific input on the NARMS program from panel of experts in microbiology and epidemiology. Panelists reviewed presentations by scientists involved in various NARMS activities and then provided feedback on six specific questions that were believed critical to the future success of NARMS programs. The results of this review are provided in the Appendix II of this report.

## NARMS Internal Review Committee

In preparation for the Science Board review of NARMS, FDA convened an internal review committee with representation from FDA Center for Veterinary Medicine, Center for Drug Evaluation and Research, Office of the Commissioner, Centers for Disease Control and Prevention, and U.S. Department of Agriculture. A listing of committee members is provided in Appendix III. The Internal Review Committee was charged with conducting a self assessment and preparing recommendations for the Science Board's review.

## Four Key Areas for the Science Board Review of NARMS

NARMS is a strong program that has made excellent progress in meeting its objectives. Several years of antimicrobial resistance monitoring data have now been collected from across the United States. The NARMS program is revealing important trends in antimicrobial resistance to antimicrobial agents of importance to human and veterinary medicine. This information is helpful in identifying the source and magnitude of antimicrobial resistance and is important for the development of public health recommendations for the use of antimicrobial drugs in humans and food animals. NARMS has matured since its inception in 1996 and would benefit from the input of the Science Board on its key elements and future directions.

FDA is requesting the Science Board review four key areas within the NARMS program:

- Sampling
- Epidemiological and Microbiological Research
- Harmonization of Data Reporting
- Coordination with International Surveillance

# SAMPLING

## Introduction

Sample collection is an integral part of public health surveillance systems, including NARMS. Sampling strategies necessarily differ among the three components (arms) of NARMS and are described below. *Salmonella* was chosen as the sentinel organism for the NARMS program. *Campylobacter* was subsequently added to the program over time, as were the commensal bacteria due to their ubiquitous presence in animals, foods and humans and their potential to serve as reservoirs of antimicrobial resistance genes for bacterial pathogens.

## Retail Meat Component

Following recommendations of the World health Organization on surveillance of enteric bacteria and disease, FDA/CVM acted to expand NARMS to include sampling of meats available to the consumer at retail. A retail meat component of NARMS was necessary in order to provide additional data on the prevalence of resistant pathogens in the U.S. food supply. Prior to adding retail meat monitoring to NARMS, a year-long statistically robust pilot study was conducted throughout the state of Iowa. This study allowed CVM to evaluate methodologies, assess workload and workflow, estimate costs, and provide foundational data (prevalence, susceptibility, seasonality & *etc*.) for NARMS retail meat surveillance. Many things were learned in Iowa that were later applied to the NARMS retail meat program, such as a need to revise our log sheets for sample custody, determine which types of meats were consistently available for purchase, and refine our bacterial culture methods. Moreover, we were able to assess the frequency that a single sample was contaminated by multiple strain types, and to establish baseline contamination and antimicrobial resistance levels based on a statistically designed sampling scheme. The experience gained from the Iowa study was instrumental in the final design of the NARMS retail meat program.

The NARMS retail meat program began in January 2002 and has become the third component of NARMS. The NARMS retail meat group collaborates with the Centers for Disease Control and Prevention to conduct retail meat sampling through 10 of 11 FoodNet Sites (California, Colorado, Connecticut, Georgia, Maryland, Minnesota, New Mexico, New York, Tennessee, and Oregon). Each site purchases 40 retail meats per month (including 10 each of chicken breasts, pork chops, ground turkey, and ground beef).

From 2002-2004, a convenience sampling plan was used by the sites to purchase retail meats. Each site visited at least one store per month and purchased as many different brands of fresh (not frozen) meat and poultry as possible. Stores chosen for sampling were not revisited for at least two months. In 2005, the CVM NARMS retail meat surveillance program switched from a convenience sampling scheme to a more statistically robust randomized sampling scheme for collection of retail meats, using stores obtained from the Chain Store Guide, Tampa, FL. In preparation to this new

sampling scheme, all 10 FoodNet sites provided areas that they would sample (either counties or Zip codes). Grocery store listings were obtained via the Chain Store Guide for these areas (between 30-300 stores depending on the FoodNet site) and inappropriate stores were removed. Grocery stores were further sorted into four quadrants by latitude/longitude with the exception of California which provided three counties for sampling. For each site, we randomized the order of quadrants tested and repeated sequentially over the year. Grocery stores were further randomized within each quadrant. Five grocery stores and three back up stores were listed per month for each FoodNet site in 2005; for 2006, there are five primary and five back-up stores for each month.

Bacteria under surveillance include *Salmonella*, *Campylobacter*, *Enterococcus* and *E. coli*. All ten FoodNet sites culture the rinsate from each meat sample for the presence of *Salmonella* and *Campylobacter*. In addition, four sites (GA, MD, OR, and TN) culture all rinsates for *E. coli* and *Enterococcus*. The number of retail meats analyzed between 2002 and 2004 for *Salmonella* and *Campylobacter* are shown in Table 1, whereas Table 2 shows the number of retail meats analyzed for *Enterococcus* and *E. coli*.

**Table 1. NARMS Retail Meats Sampled for *Salmonella* and *Campylobacter***

| | 2002 (6 states[†]) | 2003 (8 states*) | 2004 (10 states[&]) |
|---|---|---|---|
| Chicken Breast | 616 | 897 | 1172 |
| Ground Turkey | 642 | 857 | 1165 |
| Ground Beef | 642 | 880 | 1186 |
| Pork Chop | 613 | 899 | 1176 |
| Total | 2513 | 3533 | 4699 |

[†] Connecticut, Georgia, Maryland, Minnesota, Oregon, and Tennessee

* California, Connecticut, Georgia, Maryland, Minnesota, New York, Oregon, and Tennessee

[&] California, Colorado, Connecticut, Georgia, Maryland, Minnesota, New Mexico, New York, Oregon, and Tennessee

**Table 2. NARMS Retail Meats Sampled for *Enterococcus* and *E. coli* (4 States[‡])**

| | 2002 | 2003 | 2004 |
|---|---|---|---|
| Chicken Breast | 390 | 477 | 476 |
| Ground Turkey | 395 | 447 | 466 |
| Ground Beef | 399 | 470 | 480 |
| Pork Chop | 390 | 479 | 478 |
| Total | 1574 | 1873 | 1900 |

[‡] Georgia, Maryland, Oregon, and Tennessee

Each participating FoodNet laboratory uses the same procedure for sample collection and microbiological analysis. Retail meat and poultry packages are kept intact until they are aseptically opened in the laboratory at the start of examination. For chicken and pork samples, one piece of meat is examined, whereas 25 g of ground product is examined for ground beef and ground turkey samples. The analytical portions from each sample are placed in separate sterile plastic bags, 250 mL of buffered peptone water is added to each bag, and the bags are vigorously shaken. Fifty mL of the rinsate from each

sample is transferred to separate sterile flasks (or other suitable sterile containers) for isolation and identification of *Salmonella*, *Campylobacter*, *E. coli*, or *Enterococcus* using standard microbiological procedures.

Once isolated and identified, bacterial isolates are sent to FDA's CVM Office of Research in Laurel, MD for further characterization including species confirmation, serotype determination for *Salmonella*, antimicrobial susceptibility testing and pulsed-filed gel electrophoresis (PFGE) analysis (*Salmonella* and *Campylobacter* only).

**Human Component**

Sampling for the human isolates is designed around the public health laboratories and is driven by the incidence of disease in humans. The human-origin isolates are sent to the CDC Antimicrobial Resistance Laboratory in the Foodborne and Diarrheal Diseases Branch in Atlanta, GA, by state and local health departments in all 50 states. State public health laboratories systematically select every 20th non-Typhi *Salmonella*, *Shigella*, and *E. coli* O157:H7 isolate submitted to their laboratory and send the isolates to CDC. *Salmonella* serotyping is performed prior to shipping at the state level. All *Salmonella* Typhi, *Listeria monocytogenes*, and non-cholerae *Vibrio* isolates are also forwarded to CDC for further analysis.

Historically, before NARMS was established, *Campylobacter* susceptibility testing began in 1989-90 by shipment of the first five sporadic isolates per month from nineteen sentinel counties across the US. In 1997 sampling began from five FoodNet sites with the submission of one isolate each week. This was expanded through the years and in 2004 included isolates submitted from ten FoodNet sites. However, the sampling scheme for selection of isolates varies by site. Since not all states require submission from clinical laboratories, some states send isolates from the state laboratory (five sites) and some from sentinel laboratories (five sites). In 2005, CDC implemented a new sampling scheme for *Campylobacter* aimed at improving the submission of a representative sample of *Campylobacter* isolates to CDC from the state public health laboratories in each of the participating 10 FoodNet surveillance sites. Instead of each site submitting one isolate per week, current sites submit a representative proportion of isolates using a systematic procedure determined by the number of isolates received per site.

CDC is also participating in surveillance aimed at determining the prevalence of antimicrobial-resistant enterococci among persons outside the healthcare setting. This study began in June 1998 with three participating sites: Oregon, Georgia, and Maryland. Since that time, the study has expanded by two sites: Minnesota Health Department and William Beaumont University in Michigan. Human stool isolates are obtained from outpatients or healthy volunteers. Enterococci are isolated and forwarded to CDC for speciation and antimicrobial susceptibility testing.

## Animal Component

The animal component collects isolates from diagnostic, on-farm and slaughter sources.

***Salmonella*:**

**Diagnostic isolates** - Diagnostic isolates from sick animals are submitted by sentinel sites, which serve as state, regional or local veterinary diagnostic laboratories and are primarily located at Universities or are collected by ARS staff from the National Veterinary Services Laboratories (NVSL)[1]. Isolates collected from NVSL are randomly selected from the database and include the same type of information obtained from the Veterinary Diagnostic Laboratories (VDLs), if available. VDL states are excluded from selection at NVSL to minimize the likelihood of duplicate isolates and only one isolate from clustered samples is selected. Animal sources include food animals (poultry, swine and cattle) as well as exotics, domestic pets and other non-food producing animals.

**Non-diagnostic isolates** – Most non-diagnostic isolates are collected either on-farm or from slaughter houses and are presumed to originate from healthy animals which enter the food chain. However, some environmental isolates and some non-diagnostic isolates from sentinel sites are also tested and included as non-diagnostic isolates.

*On-Farm Isolates*

On-Farm isolates are most often submitted from the National Animal Health Monitoring System (NAHMS)[2] studies. The USDA initiated NAHMS in 1983 to collect, analyze, and disseminate data on animal health, management, and productivity of America's domestic livestock populations. Some on-farm isolates may be submitted from other smaller, specific studies conducted by the USDA or collaborators when available.

*Slaughter Isolates*

Slaughter isolates are submitted through the USDA Food Safety and Inspection Service (FSIS) *Salmonella* HACCP Verification Testing Program (and pre-implementation testing). This program implements the 1996 Pathogen Reduction/Hazard Analysis and Critical Control Point (PR/HACCP) rule and established *Salmonella* performance standards in seven categories of raw meat and poultry products: broilers; market hogs; cows/bulls; steers/heifers; ground beef; ground chicken; and ground turkey. Isolates from baseline and ready-to-eat (RTE) sampling programs are also included for testing.

Samples submitted to NARMS are collected from all federally inspected plants throughout the United States and include carcass rinsates (chickens), carcass swabs (turkey, cattle and swine), ground products (chicken, turkey, and beef), eggs/egg products, and certain ready-to-eat (RTE) foods.

---

[1] Ames, Iowa

[2] NAHMS, USDA-APHIS, Center for Epidemiology and Animal Health, Veterinary Services, Fort Collins, Colorado, http://www.aphis.usda.gov/vs/ceah/ncahs/nahms/index.htm

There are two different categories of HACCP testing. Samples from routine testing are called "A" sets. For "A" sets, establishments are scheduled for testing approximately every six to 12 months. If an establishment fails to meet certain standards, targeted testing is conducted. The subsequent targeted testing set is called "B", followed by "C", and "D" if sets continue to fail. There are between 51 and 82 samples collected per set, depending on product class. Approximately 82 to 91% of HACCP isolates are "A" set isolates (see Table 13).

FSIS ships *Salmonella* isolates to the ARS laboratory for antimicrobial susceptibility testing. ARS sorts data by set and plant size and captures date of submission, region, animal source (chicken, turkey, swine, etc.), sample type (rinsate, swab, ground, RTE, eggs), serotype and antimicrobial resistance pattern in the database. In 2006, FSIS is considering concentrating resources at establishments with higher levels of *Salmonella*.

***Campylobacter:***

*1998-2000*

From 1998 to 2000, *Campylobacter* isolates were obtained from a variety of USDA-FSIS programs for inclusion in NARMS. In 1998, *Campylobacter* isolates were only submitted from the Eastern FSIS laboratory, whereas in 1999 and 2000, isolates were obtained from all three FSIS laboratories (Eastern, Midwestern, and Western laboratories). FSIS cultured samples for *Campylobacter* using the most probable number method described in the FSIS Microbiology Guidebook. Nalidixic acid susceptibility and cephalothin resistance were used as identification criteria for *Campylobacter jejuni/coli*. This likely resulted in an underreporting of quinolone/fluoroquinolone (Q/FQ) resistant *Campylobacter* as quinolone resistant *Campylobacter* were not submitted for testing.

*2001-Present*

Since 2001, *Campylobacter* tested in the NARMS animal component have been isolated by ARS from spent chicken carcass rinsates submitted by the Eastern FSIS laboratory as part of the *Salmonella* HACCP Verification Program. As a result of the concern over underreported Q/FQ results, ARS does not use the most probable number method described in the FSIS Microbiology Guidebook for isolation. Culture is only conducted on rinsates containing $\geq$10 ml of fluid which is then centrifuged, the supernatant is discarded and the pellet is re-suspended in media routinely used in the ARS laboratory. Use of nalidixic acid susceptibility and cephalothin resistance as a confirmatory test has been discontinued and *Campylobacter* are selected based on morphologic characteristics followed by species confirmation by PCR.

A subset of *Campylobacter* isolates have also been submitted from diagnostic and on-farm sources, but these data have not been included in the NARMS annual reports.

***Enterococcus* and *E. coli*:**

Testing for generic *Escherichia coli and Enterococcus* began in 2000. *E. coli* and enterococci are isolated by ARS from the same spent chicken carcass rinsates submitted by the FSIS Eastern Laboratory for *Campylobacter* isolation. However, all rinsates are cultured with no minimum fluid requirement. In addition, generic *E. coli* isolates may also be obtained from diagnostic and non-diagnostic sources and are not limited to food producing animals. Source (food animal versus non-food animal) is differentiated in the annual reports. *E. coli* isolates may also be obtained from environmental and other sources (e.g., fruits and vegetables).

**CDC External Review, August 12-13, 2004**

CDC conducted a formal external review of the human NARMS component in August 2004 and several recommendations were made regarding sampling strategies (Appendix I).

**NARMS Expert Review, June 23-24, 2005**

During the June 23-24, 2005 expert review, CVM asked the panel to evaluate the current sampling schemes for the NARMS program (Appendix II).

The panel agreed that a switch from a convenience to random sampling in the retail meat component was a distinct improvement. There was concern about *Enterococcus* and *E. coli* being sampled from only 4 FoodNet sites, and suggested systematic sampling from all 10 FoodNet sites (maybe 1 out of every 10 samples). Pilot studies were thought to be useful for other meats (e.g. veal, lamb or ready-to-eat meats) to determine prevalence and antimicrobial susceptibility profiles as resources permit. As *Salmonella* and *Campylobacter* are infrequently recovered from several retail meats, it was suggested that the program work with statisticians to determine effective sampling strategies to increase recovery of target pathogens.

The panel agreed that current FSIS-HAACP compliance sampling for *Salmonella* in the animal component does not yield a nationally representative sampling, but that it can provide useful information about trends. Bias may be introduced by sampling more frequently in plants that may have a *Salmonella* problem. The reviewers questioned the usefulness of including “B” and “C” FSIS compliance sets. However, they suggested a side-by-side comparison of the slaughter A, B, and C sets as well as slaughter and retail meat data to look at similarities/differences. They also thought it would be helpful to monitor antimicrobial use at farm level and to collect and test on-farm samples. The panel also agreed that use of spent poultry rinsates likely means that *Campylobacter* is less likely to be recovered due to possible temperature and time abuses and the fragility of the organism.

For all three NARMS components, it was also generally agreed that the sentinel organisms were less relevant. If forced to cut due to budget constraints, the emphasis

should be on organisms associated with human illness (*Campylobacter* and *Salmonella*). The panelists also suggested reduced testing of *Enterococcus* isolates in addition to minimizing *E. coli* testing.

**Progress on Sampling since June 2005 Review**

Implementation of several of the recommendations to improve sampling strategies among the three NARMS components has already begun and is further described below.

**Retail Component**: With regard to the NARMS retail component, the adoption of a more statistically robust randomized sampling scheme for collection of retail meats in January 2005 was seen as a very positive step to improving the program. Improvements to this sampling plan are under way as the FoodNet sites broaden their geographic areas and increase the diversity of samples collected and number of stores visited.

**Human Component**: The NARMS human component responded to recommendations made by their external review committee with regard to *Campylobacter* sampling. CDC developed a new sampling scheme, which took effect on January 1, 2005. The new strategy will be implemented in three stages with a goal of having a more robust, nationwide system. In the first stage, FoodNet sites already participating in *Campylobacter* surveillance submit a more representative sample of isolates to CDC. Instead of each site submitting one isolate per week, current sites submit isolates using a systematic procedure determined by the number of isolates received per site (see Appendix 2). This strategy results in more isolates being submitted for testing; the CDC NARMS Laboratory was able to increase its capacity for this new sampling scheme. The next stage of the new strategy, will involve the addition of non-FoodNet states that have mandatory referral of *Campylobacter* isolates. The third and final stage will include all remaining states that are willing and able to participate in the surveillance scheme. When this new scheme is realized to its fullest extent, it will eliminate many of the limitations of the current strategy that were noted by the review panel.

**Animal Component**: The ARS will initiate studies in spring 2006 to determine the validity of continued use of spent rinsate for culture of *Campylobacter*, *E. coli* and *Enterococcus*. These studies will include a comparison of shipping temperatures, hold time, removal of fluid prior to culture, and selection of multiple colonies for analysis. Until these studies are completed, culture will continue using current protocols. Further, FSIS will initiate a baseline testing study for *Campylobacter*. Since baseline studies are designed to be more robust and statistically representative, these isolates are likely to be included for testing.

With respect to concerns regarding testing 'set' and potential bias for *Salmonella* isolates, USDA is now able to sort data by set beginning in 2000; in 2005, USDA completed adding testing set information retrospectively for these years. As seen below in Table 13, the percent of isolates captured by the 'A' set ranges from 82% (in 2002) to 91% (in 2004), averaging 85% for all years. Starting in 2005, reports will include a breakdown of "A" set data, at a minimum.

**Table 3. Distribution of Slaughter Isolates* by Year and Compliance Set**

| | 2000 | 2001 | 2002 | 2003 | 2004 | Grand Total |
|---|---|---|---|---|---|---|
| Number of slaughter isolates | 3530 | 3169 | 3747 | 2456 | 2442 | 15,344 |
| Number of slaughter isolates with set information | 3480 | 3155 | 3082 | 2286 | 2426 | 14,429 |
| Number of slaughter isolates without set information | 50 | 14 | 665** | 170** | 16 | 915 |
| A Set | 85.49% | 85.10% | 81.64% | 83.84% | 90.85% | 85.22% |
| B Set | 13.71% | 13.15% | 14.60% | 10.51% | 7.13% | 12.16% |
| C Set | 0.52% | 1.68% | 3.54% | 4.77% | 1.61% | 2.27% |
| D Set | 0.29% | 0.06% | 0.23% | 0.88% | 0.41% | 0.34% |

* All isolates tested, including eggs and RTE
** The large number of isolates without set information is due in large part to egg submissions

In 2006, funding for acquisition of veterinary diagnostic isolates was discontinued as it is thought that submissions may be predominantly from animals who have failed previous antimicrobial therapy, thereby over-representing resistance phenotypes in healthy animals. However, since ill animals often co-mingle with healthy animals until removed, information obtained from testing of diagnostic isolates may serve as an early warning system with respect to identification of emerging serotypes or resistance attributes. ARS will continue to accept diagnostic isolates from sentinel laboratories until a thorough analysis of the data can be conducted. However, isolates from NVSL will not be collected.

USDA has also begun investigating how to increase and improve on-farm sampling of healthy animals instead of relying only on the FSIS isolates. This could possibly be done by enhancing and expanding the USDA Collaboration for Animal Health, Food Safety, and Epidemiology (CAHFSE) program which is currently focusing on tracking the same bacteria under surveillance in NARMS from selected swine farms to slaughter plants (see background information). CAHFSE will enable USDA to reliably track both emerging animal diseases and zoonoses within the food animal population which may affect the food supply and impact public health. However, at this time, CDC and FDA do not provide input into CAHFSE.

**Strengths and Limitations of the Sampling Program**

**Strengths:**

Retail Component

- Includes four meat types, each from a different food animal species
- Ability to develop pilot studies to investigate specific food safety issues (e.g. presence of *C. difficile* in retail meats)
- Excellent communication and coordination between CVM, CDC and participating FoodNet states
- Use of similar sampling and microbiological testing methods among the participating FoodNet states
- Linkage with other surveillance systems (FoodNet, PulseNet)

Human Component

- Expansion to nationwide representative sampling for *Salmonella, Shigella, E. coli O157*
- Improved *Campylobacter* sampling in FoodNet sites
- Excellent communication and coordination with sentinel sites
- Isolate data can be linked to other surveillance systems (PulseNet, FoodNet)

Animal Component

- Improved ability to analyze data
- Only comprehensive program for animal data in the U.S.
- Ability to conduct directed research studies to answer important questions regarding culture and characterization of isolates
- Excellent communication and coordination with sentinel sites
- Participation in NAHMS studies
- Isolate data can be linked to other surveillance systems (PulseNet, VetNet) or compared between systems (CAHFSE)

Collectively, all three components of NARMS meet either by conference call (quarterly), in small groups (as needed for directed studies, plate designs), or annually (pending availability of funding) to discuss and harmonize the three programs. Each arm is staffed by experts in their respective fields who are committed to enhancing the program to the greatest extent possible within the current budget.

**Limitations:**

Retail Component

- Limited resources and staff prevent expanding number of samples tested and/or sample collection areas
- Only certain cuts of retail meats are sampled (e.g. ground turkey compared to turkey thighs, etc.)
- Retail meat sampling is limited to 10 states, and more specifically to geographical areas surrounding testing laboratories (not a national sampling)
- Only 4 of 10 states participate in testing retail meats for *Enterococcus* and *E. coli*

- Due to decreases in NARMS funding in 2006, Maryland was removed from the program leaving 9 participating states

Human Component

- Limited resources and staff prevent expanding number of samples tested and/or sample collection areas
- Limited ability to use data to estimate burden of resistant infections

Animal Component

- Limited resources and staff prevent expansion/enhancement of program
- Slaughter isolates are from a regulatory compliance program that is not designed to yield a nationally representative sample
- FSIS will likely be moving to a more risk-based approach to *Salmonella* testing which will result in a more biased sample of slaughter isolates
- On-farm isolates vary by source from year-to-year, making trend analysis difficult
- Difficult linking on-farm data with slaughter/processing data; hence, the implementation of CAHFSE which is under-funded as well
- No direct source of animal *Campylobacter*, *E. coli* and enterococci isolates

**Recommendations of the Internal Review Committee:**

Retail Component

- Maintain current sampling strategies for *Salmonella*, *Campylobacter*, *Enterococcus* and *E. coli*
- Continue to improve the sampling scheme by increasing the geographic area under surveillance as well as number of grocery stores sampled
- Explore strategies to recruit additional states from geographic areas that may be underrepresented (e.g. Ohio valley region)

Human Component

- Continue current sampling strategies for *Salmonella, Shigella, E. coli O157*
- Explore strategies to have states conduct more testing of isolates in order to increase sample size
- Continue *Campylobacter* sampling expansion in an attempt to have a more robust nationwide sampling scheme

Animal Component

- Continue to improve sampling scheme particularly as it relates to FSIS sampling and on-farm data
- Continue to improve collection of supplemental information in support of data analysis
- Review relevance and public health importance of testing diagnostic animal isolates and possibly consider discontinuing NARMS testing of these isolates
- Seek out represented sources of *Campylobacter*, *E. coli* and *Enterococcus* of animal origin

- Provide detailed information about the sampling scheme and source of animal isolates in annual NARMS reports
- Report results for “A” set slaughter isolates separately from results for B, C, and D sets and possibly discontinue NARMS testing of B, C, and D set isolates

## EPIDEMIOLOGICAL AND MICROBIOLOGICAL RESEARCH

### Introduction

A comprehensive research effort helps ensure that regulatory actions taken to control antimicrobial resistance will be based on sound science. This includes basic and applied research focusing on the prevalence, propagation, and persistence of antimicrobial resistant bacteria in the animal production environment and on foods of animal origin. NARMS research references cited below are listed in Appendix IV.

### Data Reproducibility and Quality Control

The dilution schemes and antimicrobial content of NARMS susceptibility testing panels has undergone several design iterations as the program has matured. This has resulted in testing arrays that now meet international standards for quality control. We also have amended the content of the panels as appropriate, to accommodate new antimicrobial entities entering the market, to omit those no longer available or of limited usefulness, or to adjust dilution ranges. The susceptibility testing panel formats undergo annual review to consider possible improvements. Customized testing panels also have been designed, and are available for use in phenotypic studies of beta-lactam and fluoroquinolone resistance.

In addition, NARMS scientists developed the first standardized *in vitro* antimicrobial susceptibility testing methods (agar dilution, broth microdilution) for *Campylobacter* (1-3) which have been sanctioned by the Clinical and Laboratory Standards Institute (CLSI/NCCLS). These methods were needed to ensure intra- and inter-laboratory reproducibility of data, which is essential to FDA's regulatory mission. Broth microdilution, which began in 2004, also amenable to semi-automation and the results are easier to interpret. Thus, the method used to test this pathogen has changed over time as validated methods became established.

### Molecular Epidemiology of Resistance Genes

NARMS phenotypes provided descriptive data on the levels and extent of antimicrobial resistance from humans, foods and animals. This data by itself is incomplete, since it doesn't indicate which of several genetic elements may underlie resistance. Characterizing transmissible resistance genes at the nucleotide sequence level provides important information on the extent to which gene transfer occurs among different bacteria, the consequences of selection pressure in the drug use environment, and the spread of resistance through the food chain. These studies are helpful for informing risk assessment models and aiding in regulatory decision making.

NARMS researchers are engaged in ongoing genetic studies to identify genes involved in antimicrobial resistance. Genetic studies focus on resistance mechanisms relevant to approved animal drugs, those conferring resistance to important classes used

in human medicine and unusual resistance phenotypes among isolates from all three NARMS sectors. Examples include:

- Identification of genes and mutations in *Salmonella* and *E. coli* conferring resistance to extended-spectrum cephalosporins (*e.g.*, *cmy-2*) and fluoroquinolones (*e.g.*, *gyrAB*, *parCE*) (4-6)
- The molecular bases of streptogramin (7,8) and high-level aminoglycoside (9-11) resistance in *Enterococcus*.
- Mutations conferring fluoroquinolone resistance and multiple drug resistance in *Campylobacter* (12)
- The gene content and distribution of integrons in *E. coli* and *Salmonella* (13-15)
- The development of PCR (16,17) and microarray platforms (18,19) for rapid isolate characterization
- Mobilization of phenotypes between pathogens and commensals

**Molecular Epidemiology of Foodborne Pathogens**

Much of NARMS molecular work is whole genome strain typing by pulsed-field gel electrophoresis (PFGE). This information is used along with susceptibility data for the purpose of assessing strain relatedness, strain source and genovar dissemination. This information also is used to populate the PulseNet database at CDC, which is used in epidemiological investigations.

NARMS isolates have been exploited as a resource to investigate molecular typing tools to help determine the animal origin of foodborne bacterial pathogens (20-22). To date, over 2000 isolates representing strains of *Salmonella* and *Campylobacter* have been characterized using a combination of two or more of the following methods: antibiotic susceptibility testing (AST); serotyping, plasmid profiling; pulsed-field gel electrophoresis (PFGE); repetitive element PCR (Rep-PCR); multilocus sequence typing (MLST); fatty acid profiling and, more recently, protein profiling. Results from serotyping, AST, PFGE and MLST have provided the following associations between animal hosts and foodborne pathogens: certain serotypes have been found to be associated only with certain food animal groups; AST profiles have shown certain resistance phenotypes to be occurring with particular animal hosts; and PFGE profiles coupled with AST profiles and MLST sequence types have been shown to occur with particular animal hosts. Protein profiling of approximately 30 isolates of one *Salmonella* serotype has identified a unique protein associated with specific PFGE fingerprint clusters. Alternative methods are being examined at the CVM Office of Research in collaboration with the Center for Food Safety and Nutrition.

**Epidemiological Studies**

Numerous important epidemiological questions arise from the NARMS data and the NARMS isolate collections. These include burden of illness estimates, case control studies, the emergence of new phenotypes and antimicrobial resistance trends. Examples of these studies include:

- Trends in antimicrobial susceptibility among *Salmonella* (23), *Campylobacter* (24) and *Shigella* (25)
- Estimates of the public health burden due to antimicrobial resistance in *Salmonella* (26) and *Campylobacter* (27)
- Identifying risk factors for *Campylobacter* infection (28)
- The epidemiology of resistance in rare *Salmonella* serotypes (29)
- The emergence of ceftriaxone resistance (4,30)

**Historical Trends**

NARMS data extend back only to 1996. To better interpret current antimicrobial resistance levels, CVM contracted with the American Type Culture Collection (ATCC) to measure resistance among historical isolates of *Salmonella*, *Campylobacter* and *E. coli*. Testing is completed for of over 1,100 *Campylobacter*, 1,500 *E. coli* and 1,800 *Salmonella* strains, the latter of which date back as far as 1948. Data from this study will help us better understand NARMS data by providing "pre-NARMS" susceptibility levels and trends over the past six decades of antimicrobial use in veterinary and human medicine.

**Impact of Antimicrobial Use in Farm Animals**

To bolster NARMS data used to support FDA risk assessment models, and to better infer causes of resistance, studies are conducted to evaluate the impact of antimicrobial use on the evolution of resistance in foodborne bacteria in the target food animal species. These include on-farm studies (31-38) and studies using experimental groups of animals (1,39). For example, NARMS data indicated a rise in fluoroquinolone resistance in *Campylobacter* (24). This prompted research designed to directly measure the impact of fluoroquinolone use in broilers (23), a major reservoir of *Campylobacter*. This research was instrumental in supporting CVM's regulatory action in removing fluoroquinolones from use in poultry. These targeted studies also show how research is a by-product of the NARMS program, and is needed to fully evaluate NARMS phenotypic data.

**Use of NARMS Data in the Review of New Animal Antimicrobial Drug Applications**

One way in which CVM achieves its mission goals is through policy and guidance. In 2003, FDA/CVM updated its regulatory policy to include an antimicrobial resistance risk assessment requirement (in addition to demonstrated safety and efficacy requirements), for all antimicrobials used in food-producing animals, with the publication of Guidance for Industry #152 (see background information). This policy recognizes the potential threats to antimicrobial effectiveness in human medicine posed by antimicrobial use in food animals. Sponsors of antimicrobial new animal drugs can use NARMS susceptibility data, in addition to that from other surveillance programs or studies, when conducting safety assessments. This information primarily provides CVM with an estimate of the magnitude, or baseline, of resistance to the drug or drug class of interest among selected foodborne pathogens and commensal bacteria.

**CDC External Review, August 12-13, 2004**

CDC conducted a formal external review of the human NARMS component in August 2004 and several recommendations were made regarding research activities (Appendix I).

**NARMS Expert Review, June 23-24, 2005**

CVM presented past research projects in antimicrobial resistance and asked the panel for input on how these efforts support NARMS (Appendix II). The experts were very supportive and acknowledged the importance of research that could be linked with epidemiological information so as to be relevant to protecting public health.

It was recommended that all NARMS isolates be analyzed in a timely fashion and available to everyone for interpretation; and that the interpretation be done in concert with the three entities and with discussion. It was noted that the PulseNet program would be enhanced by integrated sampling between the three components.

Research studies should be relevant to human health outcome; not just looking at resistance and treatment failure, but also examining potential links between resistance and virulence of pathogens. It was suggested that more attention be placed on understanding of the role of commensal populations as reservoirs of resistance determinants, and determinations of the amount and type of transfer of genetic determinants from commensals to human pathogens (including non-enteric pathogens).

It was suggested that more effort be dedicated to developing and evaluating new techniques, such as microarrays. The panel felt that the use of microarrays can help speed up the detection and characterization of resistance and virulence genes.

**Strengths and Limitations of NARMS Research**

**Strengths:**

- Expertise and dedication of staff members from all arms
- Cooperative integration of FoodNet and retail meat sampling
- Consensus approach to development of antimicrobial testing panel formats

**Limitations:**

- Funding. While we understand that research is a vital part of the NARMS program, personnel and material costs for NARMS related research are not part of the NARMS budget. Each site must find separate funds to support this work.
- Personnel limitations. Due to normal turnover and heavy workloads, any reduction is staff would lead to the elimination of some current functions.
- Antimicrobial use information. In evaluating NARMS genotypes, phenotypes and the impact of mitigation actions, it is important to know the

extent of antimicrobial selection pressure in different environments. This would allow one to link human drug-use data to clinical information (e.g., diagnosis, severity of illness, and outcome), link agricultural drug-use data to species and usage patterns, and assess potential effects of geographic variations in drug use on the incidence and prevalence of antimicrobial resistance. This goal is a top priority in the federal interagency Public Health Action Plan to Combat Antimicrobial Resistance (http://www.cdc.gov/drugresistance/actionplan/)

**Recommendations of the Internal Review Committee:**

- All NARMS partners should continue to investigate emerging resistance phenotypes of human and veterinary importance
- Coordinate research between all NARMS components including joint publications in peer reviewed scientific journals
- Investigate and evaluate new technologies for characterizing resistant strains at the molecular level

## HARMONIZATION OF DATA REPORTING/RECORDING

### Introduction

Currently each NARMS component prepares a separate comprehensive annual report on their respective websites, which are linked on the FDA website.

FDA (http://www.fda.gov/cvm/narms_pg.html#Data)
CDC (www.cdc.gov/ncidod/dbmd/narms)
USDA (http://www.ars.usda.gov/Main/docs.htm?docid=6750)

FDA began publishing retail meat annual reports in 2002. Reports include prevalence information, percent resistance, multidrug resistance, MIC distributions to individual antimicrobial agents, and PFGE (*Salmonella and Campylobacter* only*).*

USDA has published an annual report since 1997. The report includes information on the clinical status (diagnostic versus non-diagnostic which includes both on-farm and slaughter/processing), source by animal species, and *Salmonella* serotype (including phage type where appropriate). Results are presented as percent resistance/multi-drug resistance and minimal inhibitory concentration (MICs) distributions.

CDC has published reports since 1996. These reports are similar in format from 1997-2002. The format will change for the 2003 annual report (currently in clearance) which will be released in March of this year. The CDC annual reports include tables showing MIC distributions, yearly summaries of percent resistance by organism/serotype/species, and yearly summaries of percentage of all isolates showing multidrug resistance and common phenotypic resistance patterns. Tables are also presented on trends of resistance over time, and logistic regression analysis for significant trends is performed where applicable.

NARMS scientists previously met in Athens, GA in 2004 and 2005 to discuss the steps needed for integration and harmonization of data and reporting. The meetings centered on common areas for focused collaboration, development of complementary data management systems and analysis algorithms, timelines, and harmonization of the annual reporting formats and publications.

### CDC External Review, August 12-13, 2004

CDC conducted a formal external review of the human NARMS component in August 2004 and several recommendations were made regarding data reporting (Appendix I).

### NARMS Expert Review June 23-24, 2005

During the June 23-24, 2005 Expert Review, the panel was asked to evaluate the current reporting system for NARMS data (Appendix II). They recommended that reporting between the three arms should be harmonized so that the data is presented in

comparable formats to facilitate data comparisons and analysis between the three arms. They recommended that this harmonized report format should be implemented as soon as possible.

The panel also recommended that a combined report from the three arms, in the form of an executive summary, be compiled. This summary report should be done no later than April of each year and include the following:

- A table comparing the top 10 serotypes from each arm, reported as a percent of the total number of isolates tested for the year
- A table showing the top 10 serotypes in people and the matching serotypes found in the animal and retail arms, reported by species or meat type
- A percent SIR (susceptible, intermediate, resistant) table for each serotype by antimicrobial comparing all years
- A MIC (minimum inhibitory concentration) table by serotype by antimicrobial by state (human), source (animal) and food (retail meat) for the current year and for selected antimicrobials for multiple years

The panel also advocated that the data be reported more quickly and suggested that these reports be completed within 6 months after the end of the calendar year as resources permit. The panel also noted that the reports could be used to argue for prudent use of antimicrobials in humans and food animals. Examples of proposed tables and figures for the NARMS integrated report can be found in Appendix V.

**Strengths and Limitations of Data Reporting**

**Strengths:**

- Staff from all arms are committed to combined reporting and harmonization of databases
- Current advances in database development are being shared among all arms and adopted by each arm as quickly as possible
- In 2006, a jointly agreed upon executive summary is planned and will be published in the MMWR or other suitable journal
- Examples of how NARMS reports have been used:
    a. Stakeholders used the retail meat data in the "Tulathromycin Solutions for Parenteral Injection For Treatment of Bovine and Swine Respiratory Diseases Microbiologhical Effects on Bacteria of Human Health Concern A Qualitative Risk Estimation" (http://www.fda.gov/cvm/Documents/Tulathromycin.pdf )
    b. NARMS human data has been used to support and tailor outbreak investigations associated with particular antimicrobial resistance profiles.

**Limitations:**

- Dedicated IT personnel and funding to maintain, update, and provide web access to databases
- Different clearance processes exist for each agency which results in staggered annual report releases
- Little coordination among the various annual reports which makes meeting the needs of all stakeholders difficult
- Dependent on site submission of isolates which presently makes timely reporting very difficult
- Lack of access and/or availability of data on antimicrobial use in human medicine, agriculture, veterinary medicine, and consumer products that is needed to complete an integrated report.

**Recommendations of the Internal Review Committee:**

- All agencies should continue to publish separate annual reports while moving towards harmonization of report format and content
- Publish a yearly executive summary which combines data from all three components
- Where appropriate include data generated from international partners, including Canadian Integrated Program for Antimicrobial Resistance Surveillance (CIPARS) for comparison; alternatively, provide links to these other reports and include a synopsis of significant findings
- Develop web-based custom reporting/querying features
- Combine PFGE analysis with NARMS data in future annual reports

## COORDINATION WITH INTERNATIONAL SURVEILLANCE

### Introduction

Food safety is an international issue; the global trade in food products has escalated enormously over the last decade and is expected to continue to grow. Likewise, antimicrobial resistance is a global problem and foodborne diseases and resistance need to be addressed globally.

Continuous monitoring of antimicrobial resistance in enteric pathogens in both human and animal populations is established in Canada, Denmark, France, the Netherlands, Norway, Sweden and the United States (NARMS). Several other countries periodically monitor for resistance. The surveillance systems share several similarities, many by design. Since its inception, NARMS staff has collaborated with several of these antimicrobial resistance monitoring systems. These are summarized below.

### Global Salmonella Surveillance (WHO-GSS)

WHO began a program of Global Salmonella Surveillance (WHO-GSS) in January 2000, which consists of a global network of laboratories and individuals involved in surveillance, isolation, identification, and antimicrobial susceptibility testing of *Salmonella*. The program rapidly expanded to include other enteric foodborne pathogens. The objectives of GSS are to strengthen the capacities of national and regional laboratories in the surveillance of *Salmonella*, other major foodborne pathogens, and antimicrobial resistance in *Salmonella* and *Campylobacter* from humans, food and animals and to contribute to the global effort of containment of antimicrobial resistance in foodborne pathogens. As its primary activity, GSS conducts regional training courses on foodborne disease surveillance and resistance testing, it also runs external quality assurance programs on *Salmonella* serotyping, *Campylobacter*, and antimicrobial susceptibility testing, an Electronic Discussion Group, reference testing of selected *Salmonella* strains, and develops regional centers of excellence and training sites.

NARMS staff from FDA-CVM and CDC play integral roles as steering committee members for the WHO-GSS program. The steering committee consists of public and veterinary health agencies from around the world. NARMS staff serves as organizers, trainers and consultants for WHO-GSS international training courses, in both basic and advanced microbiology and in integrated microbiology and epidemiology courses. Approximately 10 countries attend each training course, which are held regionally every 12-18 months. The courses last 5-6 days and have been enormously successful in developing the practical skills of the participating microbiologists and epidemiologists, in encouraging interaction between the disciplines, and in developing future plans of action for each country. WHO-GSS also has succeeded in helping regional centers assume ongoing responsibility for educating and training regional public health authorities in foodborne disease surveillance and outbreak response. They have also helped both regions and individual countries develop focused projects in foodborne

disease and antimicrobial resistance surveillance. In this way, the activities of WHO-GSS have been important in influencing permanent improvements in global health.

**INISAR (International Network of Integrated Surveillance for Antimicrobial Resistance for Enteric Bacteria)**

Efforts to communicate and collaborate with successful integrated surveillance systems around the world are important for the NARMS program as it expands and matures. In the past, this has mainly consisted of interaction in informal settings at international conferences or meetings. In 2004, NARMS staff began developing the concept of forming a communication network, which would consist of people working with integrated surveillance systems around the world. In 2005, a meeting was hosted by Health Canada and the group was officially formed. The network is currently called INISAR (International Network of Integrated Surveillance for Antimicrobial Resistance for Enteric Bacteria) and CDC NARMS staff moderates the electronic discussion group.

**PulseNet International**

PulseNet is a national network of public health laboratories that perform DNA "fingerprinting" on foodborne bacterial pathogens. The network permits rapid comparison of these "fingerprinting" patterns through an electronic database at the CDC. The DNA "fingerprinting" method is called Pulsed-field gel electrophoresis (PFGE). PulseNet was established in 1996 through a collaborative effort of CDC, FDA, USDA and state health departments. PulseNet has been successfully to detect foodborne disease clusters, facilitate early identification of common source outbreaks and assist epidemiologists in investigation during outbreaks. The PFGE method has rigid standardization and quality assessment/quality control/certification. A national databank of PFGE patterns is maintained at CDC. FDA/CVM is the only institution submitting information on strains from food animals and retail meats.

PulseNet is moving to become an international standard. PulseNet Canada has had all provinces participating since 1999, with real-time sharing of PFGE patterns and information. PulseNet Europe currently consists of 29 countries and 54 institutes funded by the EC. PulseNet Asia-Pacific has 10 participating countries, with a training workshop held in February 2004. PulseNet China is beginning to participate, following a training workshop in September 2004; and PulseNet Latin America has over 12 countries participating since July 2004.

**Resistvet**

Since 2001, NARMS staff has collaborated with medical microbiologists in four agricultural states in Mexico and established an antimicrobial resistance monitoring system called Resistvet. The participating states are Yucatan, San Luis Potosi, Michoacan, and Sonora. Isolates of *Salmonella* and *E. coli* are collected from clinically ill humans, healthy asymptomatic children in kindergartens, and poultry and pork from

markets and supermarkets in the area of the sampled kindergarten. Training of personnel in isolation, identification and susceptibility testing was conducted during the first year of the project. Results from the Resistvet project on risk factors for quinolone-resistant *E. coli* in Mexican children and on the emergence and dissemination of extended spectrum cephalosporin-resistant *Salmonella* Typhimurium have been published. FDA/CVM no longer monetarily supports this project but NARMS staff continues to collaborate with the Mexican investigators and provides technical assistance. Several targeted research studies have evolved from Resistvet activities, which have important implications for public health in Mexico. The Mexican Ministry of Health is providing funding for the continuation of Resistvet.

**NARMS Expert Review, June 23-24, 2005**

CVM presented the involvement of NARMS in global efforts in health and food safety relating to antimicrobial resistance surveillance to the Expert Review Panel and asked the panel whether and how NARMS should continue to be involved. The experts were very supportive of the NARMS international involvement and FDA/CVM support and recommended that it continue. The three issues that NARMS has focused on, training scientists worldwide, standardization of testing methods, and standardization of reporting were recognized by the panel as critical to global efforts to control emerging foodborne pathogens and antimicrobial resistance.

The expert from WHO commented that NARMS is very important to their program on examining antimicrobial resistance arising from antimicrobial use in food animals and that integrated surveillance will help speed the development of antimicrobial resistance management strategies. It was also recommended that NARMS experts continue with international training and consultation on laboratory methods development and quality control. The panel felt that one of the legacies of NARMS will be its international extension, especially in helping establish surveillance programs in developing countries.

**Strengths and Limitations of International Coordination and Surveillance**

**Strengths:**

- NARMS involvement in international surveillance programs and global public health efforts in food safety is well established and beneficial to the U.S. consumer
- Staff are committed to collaborating with international partners
- Leadership in global programs and initiatives seen as essential

**Limitations:**

- Resources for international work in this area are very scarce

**Recommendations of the Internal Review Committee:**

- We plan to continue our involvement in all the activities outlined to the extent that funding is available.

## QUESTIONS FOR THE FDA SCIENCE BOARD

NARMS is a robust program and an important part of national public health surveillance in the United States. It has broad support from diverse sectors and numerous stakeholders. Because FDA regulates the use of antimicrobials and other drugs in animals and humans and responds with appropriate action if public health is threatened, it is vital that NARMS be based on scientifically sound foundations. Following our recent expert reviews and internal assessment, we request the FDA Science Board address four specific questions relevant to the continued success of the program:

1) **Are there inherent biases in the sampling strategies employed in NARMS? If so, how can they be improved to ensure that the data and our interpretation are scientifically sound given current resources?**

2) **Are there epidemiological and/or microbiological research studies that would better serve the goals of NARMS and the regulatory work of FDA?**

3) **Are our current plans for data harmonization and reporting appropriate? If not, what would you consider the top priorities for advancing harmonized reporting?**

4) **Are the current NARMS international activities adequate to address the worldwide spread of antimicrobial-resistant foodborne bacteria?**

**APPENDIX I**

# NARMS External Review (Aug 12-13, 2003) Document with Responses from the Centers for Disease Control and Prevention

CDC Responses in Highlighted Boxes

Foodborne and Diarrheal Diseases Branch
National Center for Infectious Diseases
Centers for Disease Control and Prevention

NARMS External Review Panel:

1. Timothy Jones, MD
2. Scott McEwen, DVM, DVSc
3. Dale Morse, MD, MS
4. David Paterson, MD, PhD
5. Lyle Vogel, DVM, MPH
6. Patricia Winokur, MD

## I. Introduction and Overview:

A panel of outside experts met at CDC on August 12-13 to review the NARMS Program. Prior to completing this report, the panel reviewed a draft of the Centers of Disease Control and Prevention's 2002 NARMS Annual Report and other NARMS publications, and listened to presentations from NARMS scientists and staff followed by discussion of related issues.

The National Antimicrobial Resistance Monitoring System (NARMS) combines the activities of the U.S. Food and Drug Administration (FDA), the Centers for Disease Control and Prevention, and the U.S. Department of Agriculture (USDA) to create a nationwide monitoring system. As a part of NARMS, isolates of foodborne bacteria including *E. coli*, *Salmonella*, *Enterococcus*, and *Campylobacter* from humans, retail meats, and animals are collected and tested to monitor changes in resistance to antimicrobial drugs. The human samples for NARMS are collected from sick people and tested by CDC. The human samples are sent to the CDC National Center for Infectious Diseases in Atlanta, Georgia by participating state and local health departments. The animal samples are gathered from healthy farm animals, animal clinical specimens, carcasses of food animals at slaughter, and ground products at processing plants and are tested by USDA. Bacterial isolates of animal origin are collected from sites across the U.S. and sent to the Agricultural Research Service Antimicrobial Resistance Research Unit of USDA in Athens, Georgia. Animal isolates also come from federally inspected slaughter and processing facilities, USDA's animal health monitoring studies on farms, and veterinary diagnostic laboratories. The retail meat samples are collected from grocery stores in states participating in the Foodborne Diseases Active Surveillance Network. Participating laboratories from FoodNet states isolate bacteria of interest and forward the isolates to the FDA Center for Veterinary Medicine Office of Research Laboratory in Laurel, Maryland for further analysis.

The principal objectives of NARMS are:

- Monitor antimicrobial resistance among bacteria that cause intestinal infections,
- Provide a platform for studies to determine factors contributing to resistance and public health impact of resistance (e.g. field investigations, case-control studies),
- Guide intervention efforts to mitigate antimicrobial resistance.

NARMS is a very strong program that has made some excellent progress in meeting these objectives. Several years of antimicrobial resistance monitoring data have now been collected on important human pathogens (e.g., *Campylobacter*, *Salmonella*, *Shigella*) and commensals (e.g. enterococci) that were collected from across the U. S.. This is a very valuable resource that is revealing important trends in resistance and decreased susceptibility, as well as increased susceptibility, to a wide variety of antimicrobial agents of importance to human and veterinary medicine. NARMS data have been used to support public policy and were critically important for the recent FDA-CVM quantitative assessment of risk to human health from fluoroquinolone resistance in *Campylobacter*. There will be a continuing need for high-quality, population-based antimicrobial resistance data to support regulatory and other science-based efforts to mitigate

antimicrobial resistance. Furthermore, NARMS is highly regarded internationally (e.g., Canada modelled much of its antimicrobial resistance monitoring program (called CIPARS) on NARMS) and NARMS staff contribute to international monitoring and capacity through WHO expert consultations on antimicrobial resistance and Global Salm-Serv.

## II. Sampling strategy for NARMS *Campylobacter* isolates

### A. Background

Historically, before NARMS was established, *Campylobacter* susceptibility testing began in 1989-90 by shipment of the 1st five sporadic isolates per month from nineteen sentinel counties. In 1997 sampling began from five FoodNet sites with the submission of one isolate each week. This was expanded through the years and for 2004 includes isolates submitted from ten FoodNet sites. However, the sampling scheme for selection of isolates varies by site. Because not all states require submission from clinical laboratories, some states send isolates from the state laboratory (five sites) and some from sentinel laboratories (five sites).

### B. Questions

#### 1. What are the strengths and weaknesses of the current sampling strategy for *Campylobacter*?

Strengths:

- Convenience sample
- Low cost
- Tied to FoodNet site data
- Already up and running
- Several years data available to follow trends

Weaknesses:

- Not generalizable/representative
- Not related to population or incidence
- Different sampling methods from 10 participating sites (5 utilize samples sent to state lab, 5 utilize samples from sentinel labs)
- Not getting the target number of isolates per site (e.g., average of only 35 isolate per site in 2002 versus target of 53, one per week)
- Not exploiting potential of sampling from 12 states with mandatory referral (though not sure of completeness of referrals)

- Now utilizing E-test which is labor intensive and limits potential number of isolates that could be tested (up to 700 a year)
- Really need better quality data to withstand criticism of validity of data to address status of fluoroquinolone resistance

We agree with the assessment of the NARMS External Review Panel regarding the strengths and weaknesses of the current *Campylobacter* sampling strategy. It is important to improve the *Campylobacter* sampling strategy resulting in a more representative sample. We have made changes to the *Campylobacter* sampling scheme, as explained below, to address this issue raised by the review panel.

**2. Should the sampling strategy for *Campylobacter* be changed? If so, how?**

*Campylobacter* susceptibility testing is a high priority and needs to be maintained. However, the limitations of the current sample need to be examined/evaluated because of the lack of generalizability and lack of representativeness.

Recommendations:

- Since the limit on the number of samples that can be tested per year (e.g., 700) in itself creates some of these limitations, CDC needs to examine methods that could be used to increase the number of specimens tested (e.g., change in laboratory methods, development of standardized procedures that could be utilized in the field by partner sites instead of relying solely on CDC, redirection of resources within NARMS to increase sample size, etc.)

- CDC should systematically compare the merits of the current system and an alternate option (e.g., utilization of 12 states with mandatory referral of isolates or utilization of large national laboratories) using a side by side analysis of major features (completeness, geographic distribution, ability to audit, etc.). If the current system is retained, changes should be made to increase completeness of submission and representativeness (e.g., taking into account sampling by population and incidence as well as by time of year, improving percentage of isolates received per site per week, etc.).

- If substantial changes are not made, we encourage considering a one-time "validation" study of the current system. Devotion of resources to a more intensive survey of specimens from a larger and more diverse population for a limited time, would allow demonstration of the validity of the current system (or identify specific problems for correction).

The NARMS External Review Panel suggested changing the sampling strategy to incorporate additional sites and develop a more comprehensive approach within the current sites. In response, we have developed a new sampling scheme, which took effect on January 1, of 2005. The new strategy will be implemented in three stages with a goal of having a more robust, nationwide system. In the first stage, we are working with the FoodNet sites already participating in the *Campylobacter* surveillance to receive a more representative sample of isolates. Instead of each site submitting one isolate per week, current sites have begun submitting isolates using a systematic procedure determined by the number of isolates received per site [Table 1]. This new strategy will result in more isolates being submitted for testing; the NARMS Laboratory has made the necessary efforts to increase its capacity in preparation for this new sampling scheme. The next stage of the new strategy, planned to go into effect in 2006, will involve the addition of non-FoodNet states that have mandatory referral of *Campylobacter* isolates. The third and final stage will include all remaining states that are willing and able to participate in the surveillance scheme. When this new scheme is realized to its fullest extent, it will eliminate many of the limitations of the current strategy that were noted by the review panel.

## III. Reporting and Dissemination of Data on Susceptibility Testing

### A. Background

Currently NARMS prepares a comprehensive annual report with a large quantity of data, which is widely disseminated. The NARMS Program has recently made marked improvements in its website, and monitoring indicates that this website receives a large number of visitors from a spectrum of agencies and backgrounds. Reports are available for a number of years, which allows comparison of data over time.

### B. Questions

**1. What are the advantages and disadvantages of reporting data as percent resistance vs. percent non-susceptible?**

Currently much of the summary data is reported in terms of percent resistance. In the narratives, intermediate resistance is referred to but "non-susceptible" is not generally addressed by NARMS.

Recommendations:

From a clinical and molecular perspective, presenting results primarily in terms of "non-susceptible" compared with susceptible is the most useful. However, we advocate continuing to present the basic data in tables and figures in a variety of formats. This would allow users/readers to interpret data using a variety of criteria or different labels. We recognize that this change will present challenges in terms of comparison of future reports with earlier ones, but feel that this will be accepted and understood by users.

We agree with the NARMS External Review Panel that reporting results in terms of "non-susceptible" and susceptible is useful in many instances. We are in the process of reviewing how NARMS' international peers report their findings for enteric bacteria in order to gather more data on the issue. Our current plan is to include both forms of results in future reports. For the NARMS 2003 Annual Report, the summary results will be presented as both resistant and intermediate.

**2. How should NARMS improve the annual report?**

Recommendations:

Given the importance of NARMS data, efforts should be made to disseminate reports in a timely manner. Currently the 2002 annual report is expected to be completed by the end of 2004. It seems reasonable to make efforts to complete and distribute a final summary of data within one year (i.e., the 2004 Annual Report would be complete by December, 2005) and possibly the tables and figures could be posted on the CDC website within 6 months (i.e., the tables for 2004 could be posted in July 2005). We recognize that substantial efforts have already been made along these lines, and encourage continued attention to timeliness.

The current annual report is large and very detailed, and available in print and on the website in its entirety. We recommend that a "Summary Report" be prepared, using the DANMAP 2003 annual report as a model, which will include summary tables and figures interspersed with narrative in a polished printed format, which would also be available for download on the website. Detailed tables and figures (in a variety of permutations)

would be available on the website, to which readers would be directed. We recommend that the website be developed to allow for searching and interactive queries by the user, to answer specific questions.

The current annual report has a huge amount of detailed data, but very little narrative background. We recommend that the summary Annual Report include a more complete "background" section that will give an overview of the complete NARMS program, including its goals, history, and collaborations with animal and retail-food programs. This should include mention of other efforts within CDC (i.e., FoodNet, PulseNet, Tenover's work, etc.) as well as outside CDC. While we realize that the current state of information-sharing with other agencies may be limited, it is important to acknowledge that other repositories of complementary data exist at FDA, USDA and other agencies, and that users should be provided with access (i.e., websites) to those resources. This might also include some "benchmarking" of NARMS data (i.e., comparison with MRL or Sentry data or other similar studies, or at least acknowledgement of other data sources and potential limitations or benefits to comparing them).

It is recommended that attention be devoted to acquiring statistical consultation to present trend data clearly and accurately (i.e., not limiting the presentation to comparing only the first and latest years' data, and not taking advantage of the wealth of additional data available).

Attention should be given to ensuring that the Annual Report is complete, and highlights all of the very nice activities in which NARMS is involved. The 2002 report, for example, makes no mention of the enterococcal resistance study or retail food study. Even if complete data are not available, it is worthwhile to acknowledge ongoing work.

It is also recommended that the summary section report susceptibles and non-susceptibles by consolidating the testing data by classes of antimicrobials. Reporting of the test results for individual antimicrobials should be retained in the tables and figures. We recognize that for trend analysis of resistance either backward conversion or continuance of reporting of resistance to each antimicrobial will be required but believe that grouping by class of antimicrobial will provide greater clinical relevance. Grouping may also increase the denominator of tests that in turn will allow greater clarity of the significance of changes in trends.

We agree with all of the NARMS External Review Panel's assessments and suggestions regarding the dissemination of NARMS data. The NARMS Laboratory has made great efforts towards testing isolates on a shorter schedule, and has already tested a high portion of the isolates received in 2004. Quicker testing combined with efforts to automate and streamline many parts of the data analysis and report generation will hopefully allow us to produce our annual reports in a more timely manner. We hope to produce the Annual Report in the summer each year with the proceeding year's data. There are also plans to present NARMS data in an issue of MMWR on a regular basis, beginning in the spring of 2006.

A committee has been formed to address many of the other issues presented by the review panel such as the tables and figures used, the summary report, and grouping by class. The committee has been meeting on a regular basis and analyzing reports from NARMS' international peers and working with the FDA and USDA components of NARMS to determine the most effective standards and means for presenting the data in future reports.

## IV. Molecular Characterization

### A. Background

The core staff members coordinating this effort have been Jean Whichard and Kathryn Gay, under the supervision of Tim Barrett (amongst others). The NARMS group has been instrumental in researching and publishing information on the CMY-2 beta-lactamase in *Salmonella* Newport. In the past, the NARMS laboratory has collaborated with prominent researchers such as Paul Fey, Alessandra Carattoli and most recently, David Hooper (to characterize plasmid-mediated quinolone resistance).

A key existing strength of the program is that the NARMS group has clearly the most comprehensive nationwide collection of human isolates of enteric pathogens in existence. There is also the potential for a collaborative effort with the FDA and USDA in molecular characterizations and comparisons with isolates of food and animal origin.

A key issue with the program has been the limitation in terms of financial resources and personnel with which to move forward with molecular characterization of this collection of organisms. A secondary issue has been the practical ability to perform molecular characterization of complementary food and animal isolates.

The NARMS group and in particular the individuals who focus on molecular testing have been very visible at national meetings and the NARMS meeting has resulted in the international dissemination of data.

## B. Questions

### 1. Should we continue to focus on ”problem” and unusual isolates?

Recommendations:

There is a unique opportunity to explore which isolates have an unusual phenotype and it is clear that the databases need to be screened on a routine basis for these unusual phenotypes (e.g., cefepime resistance, carbapenem resistance) to identify emerging problems. These are the isolates that NARMS has concentrated their molecular characterizations and our group feels that these will continue to be the most interesting and important populations on which to concentrate molecular analysis. However we do not believe that this small group of individuals who have many responsibilities and limited resources can realistically undertake the primary time-consuming identification of novel genes. Rather this task should be a collaborative effort with others within CDC or with outside experts in the field.

We agree that NARMS should continue to investigate unusual isolates, and that we should collaborate with other members of the CDC and outside experts to compensate for a lack of capacity in this area. NARMS will formalize the investigative process by establishing definitions for special isolates that warrant further examination.

### 2. Should we be more “surveillance” oriented, and what should we survey?

Recommendations:

Surveillance of new mechanisms of antibiotic resistance is an important function of NARMS and can be done within the personnel and financial constraints given. For example, if a new gene is identified in the literature that can be readily analyzed by a simple PCR analysis, NARMS isolates would be a very appropriate population that could be screened for this emerging genetic mechanism. An example might be the detection of metallo beta-lactamase genes that are likely to be rare at this time, but could emerge in the future. The other important goal would be to make sure the group utilizes the phenotypic antimicrobial resistance data effectively. The NARMS group gave an excellent example of using

quinolone resistance in conjunction with sulfonamide resistance to identify isolates that might contain a plasmid mediated quinolone resistance gene.

We agree that surveillance of new mechanisms of antibiotic resistance is an important function of NARMS and will continue to be performed.

**3. What research priorities should NARMS consider for genotype-based surveillance, applied research projects, and special studies?**

Recommendations:

The large question of how frequently to re-evaluate for known resistance mechanisms is important because this can be labor and materials intensive and not yield particularly novel data. It is not necessary to continue to evaluate QRDR mutations for *Campylobacter*, *Salmonella* and other enteric organisms since this has been well documented and is not providing new information. If there is extra time or money a periodic re-evaluation of known mechanisms might be reasonable every few years to make sure that there are no new mechanisms of resistance that are emerging, but this is clearly not a priority approach.

The use of PFGE for outbreak analysis is extremely useful. The addition of antibiotic resistance data to this is very helpful. It may not be as useful to fully characterize the genes in all outbreak isolates, but it would be useful to obtain and then analyze a small subset of the outbreak isolates by performing IEF/PCR or other techniques to confirm the genetic basis of resistance.

The suggestions given by the NARMS External Review Panel are in line with the current goals for NARMS. We are in the process of having all isolates in NARMS undergo PFGE and will continue to use PCR techniques to examine specific mechanisms of resistance.

**4. Should we emphasize special collections with extensive characterization?**

Recommendations:

A key opportunity is to perform molecular characterization for special collections of organisms that have a rich collection of epidemiological information if there is a true outcome that will be achieved. Simply

characterizing a known gene, like CMY-2, by itself could be less important, but if this data can be linked to antibiotic use, farm exposure, etc. this type of data would be extremely useful.

We agree that focusing on on special collections, with the use of epidemiological data, would be very valuable when linked with antibiotic use.

### C. Future Directions

At this point in time, direct genetic testing for antimicrobial resistance genes directly from clinical samples is not a reality. However as such technologies advance, there may be an opportunity for NARMS to collaborate with clinical laboratories to adopt this type of testing. *Campylobacter* might be an appropriate target since this is such a fragile organism, but often requires more specialized antimicrobial susceptibility testing. Also, molecular methods to replace serotyping or PFGE may arise that could expedite or improve NARMS analyses.

### D. Overall Suggestions

- We continue to support the use of molecular characterization of select NARMS isolates. The unusual or "special" isolates and those isolates that carry rich epidemiological information should continue to be the focus of molecular characterization.

- The publication of this type of material in peer-reviewed publications is essential.

- It would be very useful to see molecular characterization of the continuum of isolates from animal, food and human origins. This type of data may be useful in developing and supporting policy recommendations.

## V. Resistance in Human Commensal Bacteria

### A. Background

Antimicrobial resistance in commensals is not reported in the 2002 or earlier reports. NARMS has, however, undertaken a study of resistance in enterococci from five sites in the U.S. beginning in 1998. Its purpose was to determine the prevalence of clinically important antimicrobial-resistant enterococci in stool samples from clinic patients and healthy volunteers. NARMS recently started a prospective study of enterococci and *E. coli*, which will link epidemiological data (e.g., antimicrobial use, travel, animal contact history) with resistance data.

## B. Questions

### 1. What do you think are the most important questions to answer in this arena?

Recommendations:

It is important to monitor resistance in commensals to determine the role of these bacteria as reservoirs of resistance determinants for human pathogens. To this end, it is important, where possible, to integrate monitoring and epidemiological data (e.g., antimicrobial use) from animals, food, the environment and humans.

We agree that it is important to monitor resistance in commensals to determine the role of the bacteria as reservoirs of resistance. Efforts are underway to integrate our databases with those at FDA and USDA so that our data is in a comparable format. We agree that we need to include data on antimicrobial use in animals, food, and the environment, and will attempt to do this where the data is available.

### 2. What are the strengths and weaknesses of current NARMS surveillance for commensal bacteria?

Strengths:

- NARMS is focusing on the right commensal bacteria (enterococci and *E. coli*)
- Some FoodNet sites are being used for isolate acquisition
- Some epidemiological / demographic information on well people (volunteers) is being collected
- Convenience sampling is being employed for efficiency

Weaknesses:

- Some isolates from ill people are included, where it would be best to collect them from well people

As part of commensal surveillance we receive questionnaires that can be used to select data based on risk factors and illnesses when necessary. We believe that the convenience sampling strategy used in this surveillance is the most efficient, even though it results in samples from ill people; obtaining samples from healthy volunteers is a difficult task, however we will evaluate the potential to do so. Importantly, we do not believe that receiving samples from ill people impacts surveillance for resistance in commensal bacteria.

3. **Do you have suggestions on how to improve surveillance for human commensal bacteria of public health importance?**

Recommendations:

Where possible, it is important to access existing studies of commensals (e.g., "Unexplained diarrhea study") using standardized epidemiological and laboratory data collection. It is important to integrate the data on commensals from other branches of NARMS (i.e., animal, food and human). NARMS should focus on the important commensals (enterococci and *E. coli*) and use susceptibility panels that, at a minimum, include those drugs important to human health, especially if members of the same class are used in animals (e.g., fluoroquinolones, extended-spectrum B-lactams, glycopeptides).

We agree with the suggestions presented by the NARMS External Review Panel, and will continue to analyze our commensal data with standard epidemiological practices in line with the provided comments. We have also been in the process of re-evaluating data for commensal bacteria and retail food surveillance to make sure the data can be

## VI. General Program Recommendations

### A. Background

NARMS is an excellent program that has accomplished a huge amount with limited resources, as outlined in the introduction. During our review, we considered a number of additional comments and suggestions that are directed at further strengthening NARMS:

### B. Recommendations

NARMS would benefit from getting additional statistical review (either from within CDC or outside such as a consultative contract with a university-based

biostatistics center) of both methodology (i.e., sample size and selection criteria for all surveillance projects as well as special studies) and analyses. The latter might include an evaluation similar to what the FoodNet program has recently gone through, to address issues of doing trend analyses and summarizing data as the population under surveillance changes.

NARMS should continually re-evaluate resource allocation, to ensure thoroughly addressing questions most appropriate to this program. For example, some efforts might need to be limited (i.e., surveillance of *E. coli* O157 resistance) to improve surveillance of high-priority issues such as *Campylobacter*.

The decisions about adding or changing the panels of antimicrobials tested should be made in a systematic fashion with regular frequency. These decisions can be facilitated by seeking status as an official observer at the National Committee on Clinical Laboratory Standards meetings, where discussions occur regarding new antimicrobials, and include representatives from industry, other federal agencies, and clinicians.

Despite obstacles, we recommend continued efforts to maximize cooperation and communication between USDA, FDA and CDC to share data and protocols to maximize efficient use of resources in addressing problems of mutual interest. This should also include continuing to foster international relationships (i.e., Global Salm-Surv) as appropriate.

We encourage NARMS to aggressively publish data in the peer-reviewed literature. A large number of nice posters and talks are presented each year, and should be followed up with a proportionate number of publications.

We encourage NARMS to evaluate novel approaches to maximizing resources for surveillance testing. For example, the PulseNet model should be examined, with the possibility that selected state laboratories would develop capacity as “Centers of Excellence” to perform basic, standardized susceptibility testing, with forwarding of isolates of particular interest to NARMS for further characterization. We note that several states already perform extensive susceptibility testing, and three FoodNet states have in the past been involved in a common protocol for *Campylobacter* testing. We recognize that CDC would need to engage in extensive evaluation efforts to ensure standardization and quality control, but believe that in the long-term this investment may facilitate concentration of limited CDC resources on performance of high-level reference-lab functions.

We also encourage regular meetings, at least annually, of a “steering or oversight committee” of key representatives of all NARMS partners (physicians, veterinarians, microbiologists, consumers, animal producers, pharmaceutical industry, state health and agricultural agencies, federal

departments of health and agriculture, etc.). This should include leaders from each of the participating organizations, who can contribute substantively to discussions about projects, goals, priorities, methods, and funding and administrative issues pertaining to the long-term success of the NARMS program.

We agree with many of the recommendations made by the NARMS External Review Panel, regarding the NARMS program in general. We consult regularly with a CDC biostatistician regarding the methodology and analyses used in NARMS, and any additional statistical review would be beneficial. We also feel that it is important to allocate our resources to focus our efforts on the aspects of NARMS that are deemed the most valuable, such as the NARMS Laboratory focusing its capacity in order to test an increased number of *Campylobacter* isolates. There is a strong interest for NARMS to maximize its cooperation with its USDA and FDA components; this has already been facilitated in part by the CDC NARMS annual report committee that has been recently formed. We also agree that there needs to be a stronger push for publications and investigating novel approaches.

Two of the suggestions made by the review panel do not fully apply to NARMS. The NARMS Working Group has a committee that discusses which antimicrobials should be tested. Our system for reviewing antimicrobials is informal, but we are in the process of having a regularly scheduled review. In order to consider other suggestions regarding the overall NARMS program, discussions are underway to hold an external review of all three arms (CDC, FDA, USDA).

**Table 1: Proposed Submission Scheme for *Campylobacter*, 2005 (Updated 4/7/05)**

| State | Number of *Campylobacter* isolates received by state or sentinel laboratory* | Number of cases from FoodNet 2002 | Number of cases from FoodNet 2003 | Base estimate | FoodNet catchment population 2003 census | Proposed submission scheme** | Expected number using new scheme and base estimate | Expected number using upper limit |
|---|---|---|---|---|---|---|---|---|
| CA | 319-381 | 1018 | 871 | 319 | 3,213,848 | **1/2 (every other)** | **160** | 191 |
| CO | 180 | 347 | 371 | 29 | 2,526,245 | **1/2 (every other)** | **90** | 90 |
| CT | 169-174 | 542 | 543 | 169 | 3,483,375 | **1/2 (every other)** | **85** | 87 |
| GA | 100-120 | 664 | 622 | 100 | 8,684,715 | **all** | **100** | 120 |
| MD | 50-100 | 374 | 423 | 50 | 5,508,909 | **all** | **50** | 100 |
| MN | 850-900 | 941 | 937 | 850 | 5,059,375 | **1/5 (every fifth)** | **170** | 180 |
| NM | 64-74 | -- | -- | 64 | 1,874,614 | **all** | **64** | 74 |
| NY | 336 | 431 | 472 | 336 | 4,314,129 | **1/2 (every other)** | **168** | 168 |
| OR | 25-70 | 562 | 578 | 25 | 3,559,596 | **all** | **25** | 70 |
| TN | 5/mo | 180 | 456 | 60 | 5,841,748 | **all** | **60** | 60 |
| Total expected NARMS submissions † | | | | | | | **972** | 1140 |
| Total for analysis ‡ | | | | | | | **826** | 969 |

* Data provided by state

** First isolate for the year will be selected and subsequent isolates will be selected based on submission scheme (e.g., for every other submission, the first isolate, third, fifth, and so on will be submitted to NARMS)

† Total expected to be submitted to NARMS

‡ Used attrition of 15% for non-viable isolates, duplicates, other reasons for excluding isolates in analysis

# APPENDIX II

## NARMS Expert Review Meeting - Agenda
## June 23-24, 2005
## DoubleTree Hotel, Rockville, MD

**Thursday, June 23, 2005**

8:30 Introduction and Purpose of Meeting – **Linda Tollefson**

8:40 NARMS Background - **David White**
- Purpose
- Brief historical description and current status, i.e.:
  - Isolation procedures
  - Identification procedures
  - Antimicrobial susceptibility testing procedures
    - *Enterococcus*
    - *Salmonella* and *E. coli*
    - *Campylobacter*
      - E-test
      - Agar dilution
      - Broth microdilution
  - Evolution and current status of Susceptibility testing panels

9:10 Budget Issues – **Linda Tollefson**
- Operating Budget
- Supplies
  - Central ordering of plates
  - Service contract for ARIS
- Biennial Meetings
- Year-to-year funding

9:30 Break

9:45 Retail Meat Arm – **David White**
- History
- Sampling scheme
- Organisms studied
- Molecular studies
- Data reporting

10:15 Animal Arm – **Paula Fedorka-Cray**
- History
- Sampling scheme
- Organisms studied
- Molecular studies

Data reporting

10:45 Human Arm – **Tom Chiller/Tim Barrett**
History
Sampling scheme
Organisms studied
Molecular studies
Data reporting
Synopsis of findings from 2004 External Review
CDC Response to 2004 External Review

11:30 Questions and Answers for all presentations

12:00 Lunch

1: 00 **Specific Issues** – **Linda Tollefson**

1:05 Animal Isolate Sampling – **Neena Anandaraman/Paula Fedorka-Cray**
FSIS will present the sampling scheme for animal isolates including a discussion of the A,B,C issue.

1. E*valuate the adequacy of the current sampling process by the animal arm of NARMS taking into account the primary objective of NARMS, i.e., surveillance of antimicrobial resistance in enteric pathogens for public health.*
   *Are these samples adequate and effective for Salmonella?*
   *Are these samples adequate and effective for Campylobacter?*
   *What other pathogens should or should not be tested as part of integrated surveillance?*
   *What sources should/should not be tested?*
   *Evaluate the contribution of diagnostic isolates.*
   *Evaluate the contribution of on farm isolates.*

2:00 Retail Meat Sampling – **Terry Proescholdt**
CVM will give overview of retail meat sampling scheme, past and present.

2. *Evaluate the sampling strategies for the retail meat arm, taking into account the most recent changes. Are they adequate as currently conducted?*

3:00 Break

3:20 Reporting – **Elvira Hall-Robinson**
CVM will describe current reporting for all arms and will highlight future plans for reporting.

3. *Evaluate the current format of reporting by the three arms. Is reporting sufficient to be able to use data from the three arms effectively for public*

*health surveillance? How would you change the reports to accomplish this (human, retail, animal)? What should be retained and/or omitted in future reports?*

4:15 Molecular Characterization – **Shaohua Zhao**
CVM will give a brief summary of the importance of molecular characterization of the isolates to the NARMS program including the matching of PFGE profiles with human isolates.

4. *Evaluate whether or not NARMS has the ability to demonstrate or refute a zoonotic continuum between food borne pathogens from animal, food and humans using molecular characterization?*

## FRIDAY, June 24, 2005

8:30 Recap – **Linda Tollefson**

8:35 National/International – **Tom Chiller/Pat McDermott/Shaohua Zhao**
A description of NARMS ongoing efforts in global health and global food security via Global Salm-Surv, Pulse Net International and the National Public Health Action Plan.
5. *How should NARMS be involved in international monitoring efforts?*

10:00 Funding – **Linda Tollefson**
Recap of NARMS funding based on the discussions that have taken place.
6. *How can NARMS funding be sustained and enhanced?*

10:45 Break

***End approximately 12:00 noon***

**June 2005 Expert Review of NARMS Animal and Retail Meat Components**

The 7 external panelists for the June 2005 expert review of NARMS were:

1) Awa Aidara-Kane, PhD, Microbiologist and International Food Safety
OMS/CDS/CPE – World Health Organization
20, Avenue Appia
1211 Geneve 27, Suisse
aidarakanea@who.int

2) Sean Altekruse, DVM, PhD
Veterinarian and Food Safety Expert
Office of Policy, Programs and Employee Development
USDA/FSIS, Independence Avenue SW, Washington, DC
sean.altekruse@fsis.usda.gov

3) Susan Kotarski, PhD
Microbiologist and Food Safety Expert
Associate Research Fellow, Metabolism & Safety
Pfizer Animal Health, 7000 Portage Road
Kalamazoo, MI 49001-0199
susan.f.kotarski@pfizer.com

4) Scott McEwen DVM, DVSc, Diplomate ACVP
Microbiologist, International Food Safety and Surveillance
Professor and Graduate Coordinator
Department of Population Medicine
University of Guelph, Guelph, Ontario, N1G 2W1, Canada

5) Marissa Miller, DVM, MPH
Microbiologist and Food Safety Expert
HHS/OS, 200 Independence Ave. \0D\0AS.W.
Washington, DC 20204

6) Daniel Sahm, PhD
Microbiologist and Food Safety Expert
Focus Technologies, Inc.
13665 Dulles Technology Drive, Herndon, Virginia 20171
dsahm@focustechnologies.com

7) Lyle Vogel, DVM
Veterinarian and Food Safety Expert
Director, Scientific Activities Division, American Veterinary Medical Association
1931 N. Meacham Road, Suite 100, Schaumburg, Illinois 60173-4360
lvogel@avma.org

**<u>Six Questions Posed, June 2005 Expert Review (With Panelists Responses to Questions Summarized in Italics)</u>**

Sampling is an integral part of any public health surveillance system and necessarily differs among the three components of NARMS. Sampling for the human isolates is designed around the public health laboratories and is driven by the incidence of disease in humans.

1 **a) Sampling for the animal arm of NARMS varies by source of the isolates. For the purpose of public health surveillance, FDA prefers isolates from healthy animals presented for slaughter since these animals become food that people consume. The slaughter and processing isolates collected across the country in the animal arm only consist of *Salmonella* organisms. Is the current sampling process by FSIS adequate and effective for *Salmonella* surveillance?**

*<u>Panelists' Responses Summarized</u> - The current FSIS sampling for Salmonella is not adequate for a nationally representative sampling. Bias is introduced by sampling more frequently in plants that may have a Salmonella problem. Multiple samples are not needed - testing of B and C set is not useful. Suggest a side-by-side comparison of the slaughter and retail meat data. It would be helpful to monitor antimicrobial use at farm level and on-farm samples would be useful.*

**b) The *Campylobacter* isolates from the animal arm of NARMS come from spent poultry rinsates that were collected as part of the sampling for *Salmonella* that were sent to the FSIS lab in Athens, Georgia. Are these samples adequate and effective for *Campylobacter*? What other pathogens should or should not be isolated and susceptibility tested from the poultry rinsates? If additional sampling is suggested, please consider what we should stop doing in NARMS since no additional funding is available.**

*<u>Panelists' Responses Summarized</u> - The panelists agreed that use of poultry rinsates likely means that we are losing sensitive Campylobacter. They suggested that poultry rinsates be tested immediately and then at timed intervals to detect differences in prevalence or counts (to determine unacceptable time delays).*

*Avoid taking multiple samples from the same source. It was generally agreed that the commensals are less relevant. If forced to cut, the emphasis should be on serotypes that cause human illness (Campy & Salmonella). Suggest cutting Enterococci with minimal E. coli testing. Other pathogens should not be added – it is a very comprehensive spectrum (although C. coli in ground turkey - particularly macrolide resistant - may be worth a thought).*

2 **Sampling for the retail meat program relates to a particular product, which represents a species of food-producing animal. In 2005, the retail meat sampling methodology underwent significant change from a convenience sampling to a random sampling method. Are the current sampling strategies for the retail meat arm of NARMS adequate as currently conducted?**

*<u>Panelists' Responses Summarized</u> - The switch from a convenience to random sampling was a distinct improvement. However, there was concern about Enterococci and E. coli being sampled from only 4 FoodNet sites - suggest systematic sampling from all 10 FoodNet sites (maybe 1 out of every 10 samples). Pilot studies may be useful for other*

*meats – veal, lamb or ready-to-eat meats. Also, it may be useful to consider differential pathogen loads on skin vs. internal cuts.*

*The sampling sizes for rarer serotypes was questioned – it was suggested to utilize existing data and work with statisticians to determine effective sampling strategies for meats with lower prevalences of pathogens.*

*The routine conference calls between NARMS collaborators were viewed as being very helpful and should be continued.*

3 **Evaluate the current format of reporting the data generated in NARMS by all three components. Is reporting sufficient to be able to use data from the three arms effectively for public health surveillance? How would you change the reports to accomplish this (human, animal, retail)? What should be retained and/or omitted in future reports?**

*Panelists' Responses Summarized - It was widely accepted that executive summaries would be helpful. In the first instance, the reporting format between the three arms should be harmonized, however, a collaborative report (human, animal and retail meat – data from the 3 arms combined) should be effected as soon as possible. It was widely advocated that the goal should be to report the data more quickly (maybe 6 months). The consolidated report could be used to argue for prudent use of antimicrobials in food animals.*

*It was widely agreed that it is useful to see the MIC distributions, because as NCCLS breakpoints or susceptibility testing methods change, you can still see the changes in susceptibility. More information concerning the data (sampling strategy, possible limitations/biases, QCs, etc.) should be stated explicitly.*

*It is helpful to show data for the top 10 serotypes in humans as a referent group, then compare retail meat and slaughter serotypes to the human's top 10.*

4 **Molecular characterization of the isolates is essential to understanding the spread of resistance. What are the top three elements of a well-coordinated collaboration for the molecular characterization of isolates from all three arms so that we can demonstrate or refute a continuum from animal, food and human origins for specific pathogens and/or resistant phenotypes?**

*Panelists' Responses Summarized - It is important that the system is transparent so that it can be used by a variety of laboratories for comparisons. The methodologies should be standardized between the three arms. It is important to link the typing to other epidemiological data and to track outbreaks. It is critical that the information is available in a more timely fashion.*

*Diagnostic samples (for molecular characterization as well as other testing) were questioned – they were not as useful as representative samples. It was thought that, while PFGE and linking through PulseNet is the current molecular strategy for NARMS, microarray methods should also be pursued.*

5 **Currently, there are global efforts in health and food safety relating to antimicrobial resistance surveillance. How should NARMS be involved in international efforts?**

*Panelists' Responses Summarized - It was widely believed that the international efforts for NARMS should be continued – for very little cost, they are achieving a great deal. It was widely accepted that antimicrobial resistance is a global problem and that foodborne diseases need to be addressed globally.*

*NARMS internationally has focused on training scientists worldwide, standardization of testing methods and standardization of reporting. These three issues are recognized as critical to global efforts to control emerging foodborne pathogens. NARMS should continue efforts with existing networks, like DANMAP, CIPARS, Global Salm-Surv and ResistVet in Mexico to share information. Perhaps cultivation of various networks for funding or leveraging for NARMS (e.g., World Bank) should be pursued.*

6 **NARMS is funded by FDA through interagency agreements with CDC and USDA. For the next several budget cycles it is unlikely that funding for NARMS will be increased. How do you suggest that we enhance and sustain funding for NARMS?**

*Panelists' Responses Summarized* - I*t was thought that greater stakeholder support would be helpful to NARMS. Perhaps industry could be asked to provide funding (but true costs should be calculated to help justify requests for increased NARMS support).*

*Cutting of commensals, diagnostic sample and compliance testing should be pursued if it is necessary to cut costs but monitoring of key foodborne pathogens (Salmonella and Campylobacter) in humans, food animals and retail meat needs to be maintained. Future budget flexibility to support pilot studies would be helpful.*
*It would be useful to have each of the arms re-define their core objectives (with review and approval by the other arms). Then the arms could collaboratively develop an integrated summary of overall objectives. Finally, stakeholders could be asked to comment on these objectives (rather than a steering committee which would be time-consuming and expensive).*

## APPENDIX III

NARMS internal review committee members (affiliation for non-CVM)
Mary Bartholomew
Tom Chiller
Paula Fedorka-Cray (USDA)
Joshua Hayes
Elvira Hall-Robinson
Ibrahim Kamara
Beth Karp
Patrick McDermott
John Powers (CDER/FDA)
Jane Robens (USDA)
Gerald Rushin
Linda Tollefson (OC/FDA)
David White
Linda Youngman

## APPENDIX IV
## NARMS Research References


1. McDermott,P.F., Bodeis-Jones,S.M., Fritsche,T.R., Jones,R.N., and Walker,R.D. 2005. Broth microdilution susceptibility testing of *Campylobacter jejuni* and the determination of quality control ranges for fourteen antimicrobial agents. *J.Clin.Microbiol.* 43:6136-6138.

2. Ge,B., Bodeis,S., Walker,R.D., White,D.G., Zhao,S., McDermott,P.F., and Meng,J. 2002. Comparison of the Etest and agar dilution for in vitro antimicrobial susceptibility testing of Campylobacter. *J.Antimicrob.Chemother.* 50:487-494.

3. McDermott,P.F., Bodeis,S.M., Aarestrup,F.M., Brown,S., Traczewski,M., Fedorka-Cray,P., Wallace,M., Critchley,I.A., Thornsberry,C., Graff,S. *et al*. 2004. Development of a standardized susceptibility test for *Campylobacter* with quality-control ranges for ciprofloxacin, doxycycline, erythromycin, gentamicin, and meropenem. *Microb.Drug Resist.* 10:124-131.

4. Zhao,S., Qaiyumi,S., Friedman,S., Singh,R., Foley,S.L., White,D.G., McDermott,P.F., Donkar,T., Bolin,C., Munro,S. *et al*. 2003. Characterization of *Salmonella enterica* serotype Newport isolated from humans and food animals. *J.Clin.Microbiol.* 41:5366-5371.

5. Doublet,B., Carattoli,A., Whichard,J.M., White,D.G., Baucheron,S., Chaslus-Dancla,E., and Cloeckaert,A. 2004. Plasmid-mediated florfenicol and ceftriaxone resistance encoded by the *floR* and *bla(CMY-2)* genes in *Salmonella enterica* serovars Typhimurium and Newport isolated in the United States. *FEMS Microbiol.Lett.* 233:301-305.

6. Zhao,S., White,D.G., McDermott,P.F., Friedman,S., English,L., Ayers,S., Meng,J., Maurer,J.J., Holland,R., and Walker,R.D. 2001. Identification and expression of cephamycinase *bla(CMY)* genes in *Escherichia coli* and *Salmonella* isolates from food animals and ground meat. *Antimicrob.Agents Chemother.* 45:3647-3650.

7. Simjee,S., White,D.G., Meng,J., Wagner,D.D., Qaiyumi,S., Zhao,S., Hayes,J.R., and McDermott,P.F. 2002. Prevalence of streptogramin resistance genes among *Enterococcus* isolates recovered from retail meats in the Greater Washington DC area. *J.Antimicrob.Chemother.* 50:877-882.

8. Simjee,S., White,D.G., Wagner,D.D., Meng,J., Qaiyumi,S., Zhao,S., and McDermott,P.F. 2002. Identification of *vat*(E) in *Enterococcus faecalis* isolates from retail poultry and its transferability to *Enterococcus faecium*. *Antimicrob.Agents Chemother.* 46:3823-3828.

9. Donabedian,S.M., Thal,L.A., Hershberger,E., Perri,M.B., Chow,J.W., Bartlett,P., Jones,R., Joyce,K., Rossiter,S., Gay,K. *et al*. 2003. Molecular characterization of gentamicin-resistant enterococci in the United States: evidence of spread from animals to humans through food. *J.Clin.Microbiol.* 41:1109-1113.

10. Jackson,C.R., Fedorka-Cray,P.J., Barrett,J.B., and Ladely,S.R. 2004. Genetic relatedness of high-level aminoglycoside-resistant enterococci isolated from poultry carcasses. *Avian Dis.* 48:100-107.

11. Jackson,C.R., Fedorka-Cray,P.J., Barrett,J.B., and Ladely,S.R. 2005. High-level aminoglycoside resistant enterococci isolated from swine. *Epidemiol.Infect.* 133:367-371.

12. Ge,B., McDermott,P.F., White,D.G., and Meng,J. 2005. Role of efflux pumps and topoisomerase mutations in fluoroquinolone resistance in *Campylobacter jejuni* and *Campylobacter coli*. *Antimicrob.Agents Chemother.* 49:3347-3354.

13. White,D.G., Zhao,S., McDermott,P.F., Ayers,S., Friedman,S., Sherwood,J., Breider-Foley,M., and Nolan,L.K. 2003. Characterization of integron mediated antimicrobial resistance in *Salmonella* isolated from diseased swine. *Can.J.Vet.Res.* 67:39-47.

14. White,D.G., Zhao,S., Sudler,R., Ayers,S., Friedman,S., Chen,S., McDermott,P.F., McDermott,S., Wagner,D.D., and Meng,J. 2001. The isolation of antibiotic-resistant *Salmonella* from retail ground meats. *N.Engl.J.Med.* 345:1147-1154.

15. Zhao,S., Fedorka-Cray,P.J., Friedman,S., McDermott,P.F., Walker,R.D., Qaiyumi,S., Foley,S.L., Hubert,S.K., Ayers,S., English,L. *et al*. 2005. Characterization of *Salmonella* Typhimurium of animal origin obtained from the National Antimicrobial Resistance Monitoring System. *Foodborne.Pathog.Dis.* 2:169-181.

16. Jackson,C.R., Fedorka-Cray,P.J., and Barrett,J.B. 2004. Use of a genus- and species-specific multiplex PCR for identification of enterococci. *J.Clin.Microbiol.* 42:3558-3565.

17. Zirnstein,G., Li,Y., Swaminathan,B., and Angulo,F. 1999. Ciprofloxacin resistance in *Campylobacter jejuni* isolates: detection of *gyrA* resistance mutations by mismatch amplification mutation assay PCR and DNA sequence analysis. *J.Clin.Microbiol.* 37:3276-3280.

18. Frye,J.G., Jesse,T., Long,F., Rondeau,G., Porwollik,S., McClelland,M., Jackson,C.R., Englen,M., and Fedorka-Cray,P.J. 2006. DNA microarray detection of antimicrobial resistance genes in diverse bacteria. *Int.J.Antimicrob.Agents* 27:138-151.

19. Chen,S., Zhao,S., McDermott,P.F., Schroeder,C.M., White,D.G., and Meng,J. 2005. A DNA microarray for identification of virulence and antimicrobial resistance genes in *Salmonella* serovars and *Escherichia coli*. *Mol.Cell Probes* 19:195-201.

20. Miller,W.G., Englen,M.D., Kathariou,S., Wesley,I.V., Wang,G., Pittenger-Alley,L., Siletz,R.M., Muraoka,W., Fedorka-Cray,P.J., and Mandrell,R.E. 2006.

Identification of host-associated alleles by multilocus sequence typing of *Campylobacter coli* strains from food animals. *Microbiology* 152:245-255.

21. Foley, S. L., Walker, R. D., White, D. G., McDermott, P. F., Rhodes, B., Fedorka-Cray, P. J., Simjee, S., and Zhao, S. Comparison of subtyping methods for *Salmonella enterica* serovar Typhimurium from food animal sources. Submitted for publication. Foodborne.Pathog.Dis. 2006.

22. Harbottle, H., White, D. G., McDermott, P. F., Walker, R. D., and Zhao, S. Comparison of multi-locus sequence typing with pulsed-field gel electrophoresis and antimicrobial susceptibility profiling of *Salmonella enterica s*erotype Newport isolates. Submitted for publication. J.Clin.Microbiol. 2006.

23. McDermott,P.F., Bodeis,S.M., English,L.L., White,D.G., Walker,R.D., Zhao,S., Simjee,S., and Wagner,D.D. 2002. Ciprofloxacin resistance in *Campylobacter jejuni* evolves rapidly in chickens treated with fluoroquinolones. *J.Infect.Dis.* 185:837-840.

24. Gupta,A., Nelson,J.M., Barrett,T.J., Tauxe,R.V., Rossiter,S.P., Friedman,C.R., Joyce,K.W., Smith,K.E., Jones,T.F., Hawkins,M.A. *et al*. 2004. Antimicrobial resistance among *Campylobacter* strains, United States, 1997-2001. *Emerg.Infect.Dis.* 10:1102-1109.

25. Sivapalasingam,S., Nelson,J.M., Joyce,K., Hoekstra,M., Angulo,F.J., and Mintz,E.D. 2006. High prevalence of antimicrobial resistance among *Shigella* isolates in the United States tested by the National Antimicrobial Resistance Monitoring System from 1999 to 2002. *Antimicrob.Agents Chemother.* 50:49-54.

26. Varma,J.K., Molbak,K., Barrett,T.J., Beebe,J.L., Jones,T.F., Rabatsky-Ehr,T., Smith,K.E., Vugia,D.J., Chang,H.G., and Angulo,F.J. 2005. Antimicrobial-resistant nontyphoidal *Salmonella* is associated with excess bloodstream infections and hospitalizations. *J.Infect.Dis.* 191:554-561.

27. Nelson,J.M., Smith,K.E., Vugia,D.J., Rabatsky-Ehr,T., Segler,S.D., Kassenborg,H.D., Zansky,S.M., Joyce,K., Marano,N., Hoekstra,R.M. *et al*. 2004. Prolonged diarrhea due to ciprofloxacin-resistant *Campylobacter* infection. *J.Infect.Dis.* 190:1150-1157.

28. Kassenborg,H.D., Smith,K.E., Vugia,D.J., Rabatsky-Ehr,T., Bates,M.R., Carter,M.A., Dumas,N.B., Cassidy,M.P., Marano,N., Tauxe,R.V. *et al*. 2004. Fluoroquinolone-resistant *Campylobacter* infections: eating poultry outside of the home and foreign travel are risk factors. *Clin.Infect.Dis.* 38 Suppl 3:S279-S284.

29. Tankson,J.D., Fedorka-Cray,P.J., Jackson,C.R., and Headrick,M. 2006. Genetic relatedness of a rarely isolated *Salmonella*: *Salmonella enterica* serotype Niakhar from NARMS animal isolates. *J.Antimicrob.Chemother.* 57:190-198.

30. Dunne,E.F., Fey,P.D., Kludt,P., Reporter,R., Mostashari,F., Shillam,P., Wicklund,J., Miller,C., Holland,B., Stamey,K. *et al*. 2000. Emergence of domestically acquired ceftriaxone-resistant *Salmonella* infections associated with AmpC beta-lactamase. *JAMA* 284:3151-3156.

31. Jackson,C.R., Fedorka-Cray,P.J., Barrett,J.B., and Ladely,S.R. 2004. Effects of tylosin use on erythromycin resistance in enterococci isolated from swine. *Appl.Environ.Microbiol.* 70:4205-4210.

32. Kim,L.M., Gray,J.T., Harmon,B.G., Jones,R.D., and Fedorka-Cray,P.J. 2005. Susceptibility of *Escherichia coli* from growing piglets receiving antimicrobial feed additives. *Foodborne.Pathog.Dis.* 2:304-316.

33. Dargatz,D.A., Fedorka-Cray,P.J., Ladely,S.R., Ferris,K.E., Green,A.L., and Headrick,M.L. 2002. Antimicrobial susceptibility patterns of *Salmonella* isolates from cattle in feedlots. *J.Am.Vet.Med.Assoc.* 221:268-272.

34. Dargatz,D.A., Fedorka-Cray,P.J., Ladely,S.R., Kopral,C.A., Ferris,K.E., and Headrick,M.L. 2003. Prevalence and antimicrobial susceptibility of *Salmonella* spp. isolates from US cattle in feedlots in 1999 and 2000. *J.Appl.Microbiol.* 95:753-761.

35. Dargatz,D.A., Fedorka-Cray,P.J., Ladely,S.R., and Ferris,K.E. 2000. Survey of *Salmonella* serotypes shed in feces of beef cows and their antimicrobial susceptibility patterns. *J.Food Prot.* 63:1648-1653.

36. Englen,M.D., Fedorka-Cray,P.J., Ladely,S.R., and Dargatz,D.A. 2005. Antimicrobial resistance patterns of *Campylobacter* from feedlot cattle*. *J.Appl.Microbiol.* 99:285-291.

37. Fedorka-Cray,P.J., Ladely,S.R., Bailey,J.S., and Stern,N.J. 2001. Colonization of broiler chicks by *Salmonella typhimurium* definitive phage type 104. *J.Food Prot.* 64:1698-1704.

38. Fedorka-Cray,P.J., Dargatz,D.A., Thomas,L.A., and Gray,J.T. 1998. Survey of *Salmonella* serotypes in feedlot cattle. *J.Food Prot.* 61:525-530.

39. McDermott,P.F., Cullen,P., Hubert,S.K., McDermott,S.D., Bartholomew,M., Simjee,S., and Wagner,D.D. 2005. Changes in antimicrobial susceptibility of native *Enterococcus faecium* in chickens fed virginiamycin. *Appl.Environ.Microbiol.* 71:4986-4991.

# APPENDIX V
# Examples of Proposed Tables and Figures for NARMS Integrated Report

## Table 4. MIC Distributions for all years and all Agencies

Prepared 2-15-6; NOT FOR PUBLIC DISTRIBUTION

**Distribution of Cephalothin MICs and Occurrence of Resistance Among Non-Typhi *Salmonella* Isolates, NARMS, 1996-2003**

| | | | % of Isolates | | | Percent of all isolates with MIC (mμ/mL) of: | | | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Antibiotic | Year | N | %I | %R | CI | 0.015 | 0.03 | 0.06 | 0.125 | 0.25 | 0.50 | 1 | 2 | 4 | 8 | 16 | 32 | 64 | 128 | 256 | 512 | 1024 |
| CDC Cephalosporins I Cephalothin | 1996 | 1324 | 3.0 | **2.9** | [2.1 - 4.0] | | | | | | 0.2 | 2.3 | 53.4 | 29.1 | 9.1 | 3.0 | **1.9** | **1.1** | | | | |
| | 1997 | 1301 | 2.4 | **2.2** | [1.5 - 3.2] | | | | | | | 2.8 | 60.6 | 23.8 | 8.2 | 2.4 | **1.2** | **1.1** | | | | |
| | 1998 | 1460 | 2.2 | **2.3** | [1.6 - 3.2] | | | | | | | 1.9 | 69.0 | 20.5 | 4.1 | 2.2 | **0.8** | **1.5** | | | | |
| | 1999 | 1498 | 1.5 | **3.7** | [2.8 - 4.8] | | | | | | | 0.7 | 49.0 | 37.7 | 7.3 | 1.5 | **0.8** | **2.9** | | | | |
| | 2000 | 1377 | 1.3 | **4.0** | [3.0 - 5.2] | | | | | | | 12.8 | 60.2 | 17.9 | 3.8 | 1.3 | **0.4** | **3.6** | | | | |
| | 2001 | 1419 | 1.1 | **4.0** | [3.1 - 5.2] | | | | | | | 3.5 | 66.4 | 20.4 | 4.5 | 1.1 | **0.3** | **3.7** | | | | |
| | 2002 | 2008 | 1.4 | **5.0** | [4.1 - 6.1] | | | | | | | | 66.3 | 24.7 | 2.5 | 1.4 | **0.4** | **4.6** | | | | |
| | 2003 | 1866 | 0.9 | **5.4** | [4.4 - 6.5] | | | | | | | | 68.6 | 21.7 | 3.4 | 0.9 | **0.8** | **4.7** | | | | |
| USDA Cephalosporins I Cephalothin | 1996 | NA | | | | | | | | | | | | | | | | | | | | |
| | 1997 | 2391 | 2.3 | **2.3** | [1.8 - 3.0] | | | | | | | 1.4 | 53.2 | 34.0 | 6.8 | 2.3 | **0.7** | **1.7** | | | | |
| | 1998 | 3381 | 2.5 | **4.8** | [4.0 - 5.4] | | | | | | | 1.3 | 52.1 | 32.0 | 7.4 | 2.5 | **1.6** | **3.2** | | | | |
| | 1999 | 8508 | 2.4 | **5.3** | [4.8 - 5.8] | | | | | | | 1.6 | 54.3 | 28.6 | 7.9 | 2.4 | **1.8** | **3.4** | | | | |
| | 2000 | 7834 | 1.8 | **11.0** | [10.3 - 11.7] | | | | | | | 1.5 | 55.6 | 24.6 | 5.4 | 1.8 | **1.2** | **9.8** | | | | |
| | 2001 | 5739 | 1.7 | **13.6** | [12.7 - 14.5] | | | | | | | 3.0 | 50.9 | 25.1 | 5.8 | 1.7 | **1.6** | **11.9** | | | | |
| | 2002 | 6977 | 1.5 | **16.2** | [15.3 - 17.0] | | | | | | | | 43.9 | 31.7 | 6.8 | 1.5 | **1.0** | **15.2** | | | | |
| | 2003 | 5353 | 1.6 | **20.5** | [19.4 - 21.6] | | | | | | | | 48.2 | 24.8 | 4.9 | 1.6 | **1.2** | **19.3** | | | | |
| FDA Cephalosporins I Cephalothin | 1996 | NA | | | | | | | | | | | | | | | | | | | | |
| | 1997 | NA | | | | | | | | | | | | | | | | | | | | |
| | 1998 | NA | | | | | | | | | | | | | | | | | | | | |
| | 1999 | NA | | | | | | | | | | | | | | | | | | | | |
| | 2000 | NA | | | | | | | | | | | | | | | | | | | | |
| | 2001 | NA | | | | | | | | | | | | | | | | | | | | |
| | 2002 | 153 | 0.0 | **15.0** | (9.8-21.7) | | | | | | | | 17.7 | 56.9 | 10.5 | | **2.0** | **13.1** | | | | |
| | 2003 | 212 | 1.9 | **29.7** | (23.7-36.4) | | | | | | | | 11.3 | 46.7 | 10.4 | 1.9 | **2.4** | **27.4** | | | | |

***Notes:***
* A single vertical bar indicates the CLSI Susceptible breakpoints for each drug
* Double vertical bars indicate the CLSI Resistant breakpoints for each drug
* Unshaded areas indicate the dilution range of the Sensititre plate used to test the isolates
* Figures outside the Sensititre plate range were reported as ">" the plate's highest dilution for that drug
* 95% confidence intervals for %Resistant calculated using the Clopper-Pearson exact method

**Table 5: Percent resistance of *Campylobacter* by Agency, Species, Commodity and Antimicrobial Agent, 2002**

| Campylobacter spp. | | N | Azithromycin R | Azithromycin %R | Ciprofloxacin R | Ciprofloxacin %R | Clindamycin R | Clindamycin %R | Erythromycin R | Erythromycin %R | Nalidixic Acid R | Nalidixic Acid %R | Tetracycline R | Tetracycline %R |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| CDC | | | | | | | | | | | | | | |
| C. Coli | Human | 25 | 2 | 8.0% | 3 | 12.0% | 1 | 4.0% | 1 | 4.0% | 3 | 12.0% | 10 | 40.0% |
| C. Jejuni | Human | 329 | 6 | 1.8% | 68 | 20.7% | 6 | 1.8% | 5 | 1.5% | 70 | 21.3% | 131 | 39.8% |
| FDA | | | | | | | | | | | | | | |
| C. Coli | Chicken Breast | 90 | 7 | 7.8% | 9 | 10.0% | 7 | 7.8% | 7 | 7.8% | 9 | 10.0% | 40 | 44.4% |
| | Ground Turkey | 3 | | | 1 | 33.3% | | | | | 1 | 33.3% | 1 | 33.3% |
| | Pork Chops | 3 | 1 | 33.3% | | | 1 | 33.3% | 1 | 33.3% | | | | |
| | total | | 8 | 8.3% | 10 | 10.4% | 8 | 8.3% | 8 | 8.3% | 10 | 10.4% | 41 | 42.7% |
| C. Jejuni | Chicken Breast | 198 | | | 30 | 15.2% | | | | | 30 | 15.2% | 104 | 52.5% |
| | Ground Turkey | 2 | | | 1 | 50.0% | | | | | 1 | 50.0% | 2 | 100.0% |
| | Pork Chops | 2 | | | | | | | | | | | | |
| | total | | 0 | 0.0% | 31 | 15.3% | 0 | 0.0% | 0 | 0.0% | 31 | 15.3% | 106 | 52.5% |
| C. coli | Chicken Breast | 90 | 7 | 7.8% | 9 | 10.0% | 7 | 7.8% | 7 | 7.8% | 9 | 10.0% | 40 | 44.4% |
| C. jejuni | Chicken Breast | 198 | 0 | 0.0% | 30 | 15.2% | 0 | 0.0% | 0 | 0.0% | 30 | 15.2% | 104 | 52.5% |
| USDA | | | | | | | | | | | | | | |
| C. Coli | Chicken | 288 | 56 | 19.4% | 46 | 16.0% | 24 | 8.3% | 54 | 18.8% | 52 | 18.1% | 141 | 49.0% |
| C. Jejuni | Chicken | 526 | 5 | 1.0% | 98 | 18.6% | 4 | 0.8% | 3 | 0.6% | 120 | 22.8% | 235 | 44.7% |

**THIS DATA HAS NOT BEEN CLEARED AND IS FOR DEMONSTRATION PURPOSES ONLY**

**Comparison of data among all 3 NARMS components, 2002**

| Campylobacter spp. | | N | Azithromycin R | Azithromycin %R | Clindamycin R | Clindamycin %R | Erythromycin R | Erythromycin %R | Nalidixic Acid R | Nalidixic Acid %R | Ciprofloxacin R | Ciprofloxacin %R | Tetracycline R | Tetracycline %R |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| C. coli | | | | | | | | | | | | | | |
| USDA | Chicken | 288 | 56 | 19.4% | 24 | 8.3% | 54 | 18.8% | 52 | 18.1% | 46 | 16.0% | 141 | 49.0% |
| FDA | Chicken Breast | 90 | 7 | 7.8% | 7 | 7.8% | 7 | 7.8% | 9 | 10.0% | 9 | 10.0% | 40 | 44.4% |
| CDC | Human | 25 | 2 | 8.0% | 1 | 4.0% | 1 | 4.0% | 3 | 12.0% | 3 | 12.0% | 10 | 40.0% |
| C. jejuni | | | | | | | | | | | | | | |
| USDA | Chicken | 526 | 5 | 1.0% | 4 | 0.8% | 3 | 0.6% | 120 | 22.8% | 98 | 18.6% | 235 | 44.7% |
| FDA | Chicken Breast | 198 | 0 | 0.0% | 0 | 0.0% | 0 | 0.0% | 30 | 15.2% | 30 | 15.2% | 104 | 52.5% |
| CDC | Human | 329 | 6 | 1.8% | 6 | 1.8% | 5 | 1.5% | 70 | 21.3% | 68 | 20.7% | 131 | 39.8% |

| Table 6. Top *Salmonella* serotypes (non-typhi) tested by agency (current year) | | | | | | |
|---|---|---|---|---|---|---|
| **Rank** | **CDC** | | **USDA** | | **FDA** | |
| | **Serotype** | **Percent[a]** | **Serotype** | **Percent[a]** | **Serotype** | **Percent[a]** |
| ***1*** | Enteritidis | 52.3 | Kentucky | 42.3 | Heidelberg | 35.6 |
| ***2*** | x | x | x | x | x | x |
| ***3*** | x | x | x | x | x | x |
| ***4*** | x | x | x | x | x | x |
| ***5*** | x | x | x | x | x | x |
| ***6*** | x | x | x | x | x | x |
| ***7*** | x | x | x | x | x | x |
| ***8*** | x | x | x | x | x | x |
| ***9*** | x | x | x | x | x | x |
| ***10*** | x | x | x | x | x | x |

**[a] Percent of total number of isolates tested for the year**

**Table 7. Top *Salmonella* serotypes (non-typhi) tested from humans (CDC) and their percent distribution by animal sources (USDA) and meat commodity (FDA)[a]**

| Top Serotypes (non-typhi) from Humans | USDA | | | | FDA | | |
|---|---|---|---|---|---|---|---|
| | Chicken | Cattle | Swine | Turkey | Chicken Breast | Ground Turkey | Pork Chops |
| Typhimurium | 10.3 | 35.6 | 52.3 | 43.2 | 12.3 | 5.4 | 8.9 |
| Enteritidis | x | x | x | x | x | x | x |
| Newport | x | x | x | x | x | x | x |
| Heidelberg | x | x | x | x | x | x | x |
| Montevideo | x | x | x | x | x | x | x |
| Muenchen | x | x | x | x | x | x | x |
| Javiana | x | x | x | x | x | x | x |
| Agona | x | x | x | x | x | x | x |
| Thompson | x | x | x | x | x | x | x |
| Hadar | x | x | x | x | x | x | x |

[a] Percent of total number of isolates by each animal source or commodity

**Table 8. Ampicillin resistance among top *Salmonella* serotypes (current year)**

| Serotype | CDC | | USDA | | FDA | |
|---|---|---|---|---|---|---|
| | No. | % | No. | % | No. | % |
| **Typhimurium** | x | x | x | x | x | x |
| **Agona** | x | x | x | x | x | x |
| **Enteritidis** | x | x | x | x | x | x |
| **Heidelberg** | x | x | x | x | x | x |
| **Newport** | x | x | x | x | x | x |
| **Kentucky** | x | x | x | x | x | x |
| **Anatum** | x | x | x | x | x | x |
| **Derby** | x | x | x | x | x | x |
| **Hadar** | x | x | x | x | x | x |
| **Senftenberg** | x | x | x | x | x | x |

| Table 9. Total number of isolates tested by organism and agency (current year) | | | |
|---|---|---|---|
| | CDC | USDA | FDA |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E.coli* | x | x | x |

***OR***

| Table 10. Total number of isolates tested by organism and agency (multiple years) | | | |
|---|---|---|---|
| **1997** | **CDC** | **USDA** | **FDA** |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E.coli* | x | x | x |
| **1998** | **CDC** | **USDA** | **FDA** |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E.coli* | x | x | x |
| **1999** | **CDC** | **USDA** | **FDA** |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E. coli* | x | x | x |
| **2000** | **CDC** | **USDA** | **FDA** |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E. coli* | x | x | x |
| **2001** | **CDC** | **USDA** | **FDA** |
| *Salmonella (non-typhi)* | x | x | x |
| *Campylobacter* | x | x | x |
| *Enterococcus* | x | x | x |
| *E. coli* | x | x | x |

| Table 11. Total number of isolates tested by organism and state (CDC), animal source (USDA) and meat commodity (FDA) (current year) | | | | | |
|---|---|---|---|---|---|
| **AGENCY** | **Human** | ***Salmonella*** | ***Campylobacter*** | ***Enterococcus*** | ***E. coli*** |
| **CDC** | **Human** | x | x | x | x |
| | **Animal Source** | ***Salmonella*** | ***Campylobacter*** | ***Enterococcus*** | ***E. coli*** |
| **USDA** | Cattle | x | x | x | x |
| | Chicken | x | x | x | x |
| | Swine | x | x | x | x |
| | Turkey | x | x | x | x |
| | Equine | x | x | x | x |
| | ETC | x | x | x | x |
| | ETC | x | x | x | x |
| ***Total*** | | x | x | x | x |
| | **Meat Commodity** | ***Salmonella*** | ***Campylobacter*** | ***Enterococcus*** | ***E. coli*** |
| **FDA** | Chicken Breast | x | x | x | x |
| | Ground Turkey | x | x | x | x |
| | Pork Chops | x | x | x | x |
| | ETC | x | x | x | x |
| | ETC | x | x | x | x |
| | ETC | x | x | x | x |
| | ETC | x | x | x | x |
| ***Total*** | | x | x | x | x |

(Breakdown by State for CDC would be presented separately)

## Reporting by Region

| Table 12. Total number of isolates by region, organism and agency (current year) | | | | |
|---|---|---|---|---|
| REGION[a] | ORGANISM | CDC | USDA | FDA |
| 1 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |
| 2 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |
| 3 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |
| 4 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |
| 5 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |
| 6 | *Salmonella* | x | x | x |
| | *Campylobacter* | x | x | x |
| | *Enterococcus* | x | x | x |
| | *E. coli* | x | x | x |

[a] **Region 1= CA, NV, WA etc. etc., Region 2= SD, KS etc.**

**Table 13. Percent of isolates identified as DT104 (or any other interest group) by agency and year[a]**

| | Special interest group | CDC | USDA | FDA |
|---|---|---|---|---|
| **1997** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| **1998** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| ***1999*** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| ***2000*** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| ***2001*** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| ***2002*** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |
| ***2003*** | *DT104* | x | x | x |
| | *AKSSuT* | x | x | x |
| | *ETC.* | x | x | x |

**[a] Percent of total number of isolates tested each year by agency**
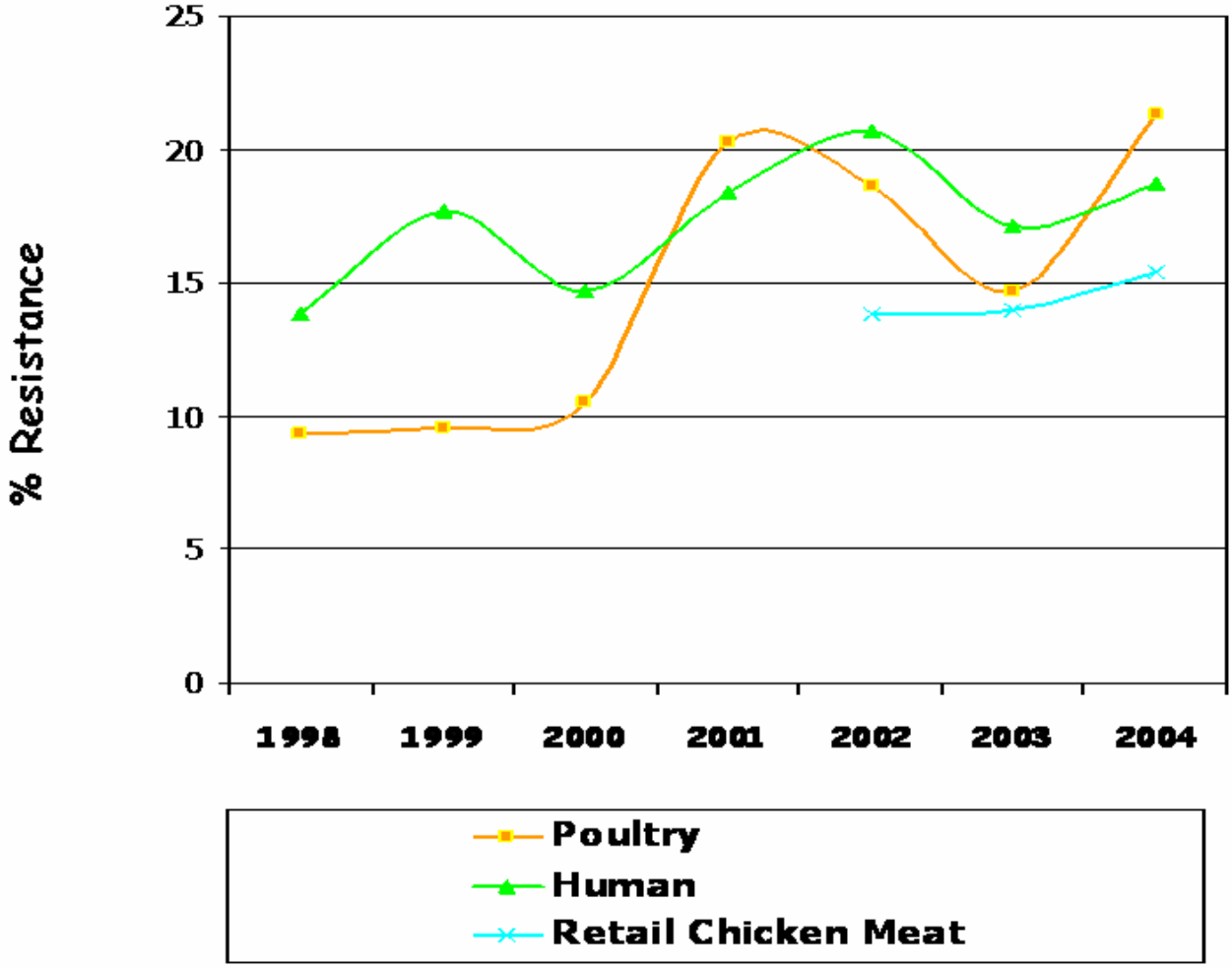
Figure 1. Ciprofloxacin resistant *C. jejuni* 1998-2004