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Quantification of antimicrobial usage in adult cows and preweaned calves on 40 large Wisconsin dairy farms using dose-based and mass-based metrics

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ABSTRACT

Use of antimicrobials in animal agriculture is under increasing scrutiny, but the quantity of antimicrobials used on large US dairy farms has not been evaluated using data from large farms and different metrics. This study investigated total antimicrobial usage (AMU) in adult dairy cows and preweaned calves (PWC) and contrasted 2 metrics used for measurement of AMU. Wisconsin dairy farms were eligible if they had >250 lactating cows, maintained computerized animal health records, and were willing to allow researchers access to treatment records. Animal health data for a 1-yr period was retrospectively collected from computerized records, and a farm visit was performed to verify case definitions and recording accuracy. Both dose-based (animal daily doses; ADD) and mass-based (total mg of antimicrobials per kg of body weight; BW) metrics were calculated at the herd, cow, and PWC levels. Descriptive statistics for AMU were examined for both age groups. Mean AMU was compared among active ingredients and route of usage using ANOVA models that included farm as a random variable. At enrollment, farms (n = 40) contained approximately 52,639 cows (mean: 1,316 ± 169; 95% CI: 975, 1657) and 6,281 PWC (mean: 180 ± 33; 95% CI: 112, 247). When estimated using ADD, total herd AMU was 17.2 ADD per 1,000 animal-days (95% CI: 14.9, 19.5), with 83% of total herd-level AMU in adult cows. When estimated using the mass-based metric, total herd AMU was 13.6 mg of antimicrobial per kilogram of animal BW (95% CI: 10.3, 17.0), with 86% of total AMU used in adult cows. For cows, 78% of total ADD (15.8 ADD per 1,000 cow-d) was administered as intramammary (IMM) preparations. In contrast, when AMU was estimated using a mass-based metric, IMM preparations represented only 24% of total AMU (12.1 mg of antimicrobial/kg of cow BW). For cows, ceftiofur was the primary antimicrobial used and accounted for 53% of total ADD, with 80% attributed to IMM and 20% attributed to injectable treatments. When estimated using a mass-based metric, ampicillin was the predominant antimicrobial used in cows and accounted for 33% of total antimicrobial mass per kilogram of BW. When AMU was estimated for PWC using ADD, injectable antimicrobials represented 79% of total usage (28.3 ADD per 1,000 PWC-d). In contrast, when AMU was estimated for PWC using a mass-based metric, injectable products represented 42% of total AMU, even though more farms administered antimicrobials using this route. When AMU in PWC was summarized using ADD, penicillin represented 32% of AMU, and there were no significant differences in ADD among ampicillin, oxytetracycline or enrofloxacin. When a mass-based metric was used to estimate AMU in PWC, oral products (sulfadimethoxine and trimethoprim-sulfa) represented more than half of the total AMU given to this group. Overall, these results showed that choice of metric and inclusion of different age groups can substantially influence interpretation of AMU on dairy farms.

Key words: antimicrobial usage, dairy, antibiotic, disease

INTRODUCTION

The discovery of antimicrobials revolutionized medicine by providing an effective method of treatment for many bacterial diseases in both humans and animals (Aminov, 2010; Davies and Davies, 2010). Many benefits of antimicrobial usage (**AMU**) have been recog-

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nized in animal agriculture, including the treatment of bacterial diseases that reduce animal welfare and production efficiency. Reduced morbidity and mortality from infectious bacterial diseases have resulted in more efficient production of animal-based protein (Johnston, 1998; Saini et al., 2012; Hao et al., 2014). Improvement in animal welfare by reducing discomfort and pain in infected animals is another important benefit.

In the United States, antimicrobial classes including aminoglycosides, lincosamides, macrolides, β-lactams, sulfonamides, and tetracyclines are used to treat dairy cows, and cephalosporins are the primary antimicrobial administered to adult dairy cows (USDA–APHIS–VS–CEAH, 2008a,b; USDA–APHIS–VS–CEAH–NAHMS, 2014). For calves, a greater variety of AMU has been reported, including tetracyclines, cephalosporins, sulfonamides, macrolides, amphenicols, and penicillins (USDA–APHIS–VS–CEAH–NAHMS, 2018). Importantly, many classes of antimicrobials are used in both animals and humans for treatment of bacterial diseases, and efforts to maintain responsible use of these antimicrobials are crucial (CDC, 2017).

Gathering quantitative data about the scope and scale of AMU on farms is an important step in understanding associations between AMU and development of antimicrobial resistance (**AMR**; Grave et al., 1999; Pol and Ruegg, 2007; Saini et al., 2012; CDC, 2014; MacFadden et al., 2016). Increased quantitative information about AMU on individual farms would help veterinarians and government agencies better understand relationships between AMU and development and transmission of AMR. Attempts to quantify AMU began in the mid-1960s in Europe with the goal of comparing AMU among countries and regions (Wade., 1984; WHO, 2003). Quantification of AMU is typically based on standardized metrics such as animal daily dose (**ADD**) or total milligrams of antimicrobial per kilogram of animal BW (Hyde et al., 2017; Mills et al., 2018).

Several studies have quantified AMU on small or mid-sized dairy farms in the United States (Pol and Ruegg, 2007; Redding et al., 2019), Europe (Stevens et al., 2016), Argentina (González Pereyra et al., 2015), and Canada (Saini et al., 2012). These studies quantified AMU using ADD with adult cows as the denominator, and estimates of usage ranged from approximately 14 to 20 ADD/1,000 cow-days. Fewer researchers have reported AMU in preweaned dairy calves (**PWC**; González Pereyra et al., 2015; Redding et al., 2019). Descriptions of AMU are useful for understanding variation in AMU among antimicrobial classes, animal categories, and diseases. However, data are lacking for AMU on larger dairy farms (>250 cows) that produce the majority of milk in the United States (MacDonald and Newton, 2014), and we are not aware of studies that have compared dose-based and mass-based metrics measuring AMU on US dairy farms. The objective of this study was to quantify AMU for cows and PWC on large dairy farms in Wisconsin contrasting dose-based (animal defined dose) and mass-based (mg of antimicrobial per kg of BW) metrics. We hypothesized that AMU would vary among farms and that use of different metrics would change the interpretation of AMU.

MATERIALS AND METHODS

Recruitment, Eligibility, and Selection of Herds

Conventional Wisconsin dairy herds were eligible for this retrospective, observational study if the farm had ≥250 lactating dairy cows when they were initially contacted, used antimicrobials to treat or prevent at least 1 event in the previous year, maintained computerized records of antimicrobial treatments, and would allow researchers access to their dairy management records.

A sampling frame of conventional dairy farms that met herd-size criteria was compiled from a list of dairy herds enrolled in previous studies (Rowbotham and Ruegg, 2015); the list had originally been compiled from dairy farm permit data obtained from Wisconsin Department of Trade and Consumer Protection (Madison, WI). The list was cross checked and supplemented from a publicly available list of Wisconsin herds classified as concentrated animal feeding operations (https://dnr.wi.gov/topic/AgBusiness/CAFO/StatsMap.html), resulting in a total of 413 potentially eligible farms. Following Institutional Review Board (University of Wisconsin-Madison, 2017–1333-CR002) approval, a postcard and a recruitment letter were mailed in June 2017. Farmers who returned a postcard indicating that they were willing to participate in the study were contacted by phone and were questioned about farm size, antimicrobial usage, and availability of records. Based on logistical and budgetary considerations as well as needs of a companion study, we sought to enroll 40 eligible farms to conduct and conclude farm visits during September to December of 2017.

Data Collection and Questionnaire

The majority of treatment records were extracted from dairy management software, but a small amount of data was retrieved from customized spreadsheets or was based on farmer recall during the onsite survey. Among dairy management software used by enrolled herds, 37 herds used Dairy Comp 305 (Valley Agricul-

tural Software, Tulare, CA), 2 herds used DairyQuest (ProfitSource, Merril, WI), and 1 farm used Afimilk (Afimilk, Fitchburg, WI).

Most of the farms that used Dairy Comp 305 sent their computerized animal health records to researchers before the farm visit. These records were reviewed to familiarize researchers with protocols and recording systems. Computerized animal health records for farms that did not use Dairy Comp 305 were obtained during farm visits. All animal health data (including information about diseases that were not treated) for adult cows (lactating and dry cows) and PWC (up to 60 d of age) were reviewed to evaluate disease definitions, detection and recording intensity, and to understand codes used for recording disease events. When using Dairy Comp 305 to obtain animal health records, a gap of 0 (all events regardless of time between episodes) was defined under ALTER\9 command to ensure that the total number of cases treated were acquired, the EVENTS\5O FOR LACT $>$ 0 command was used to obtain adult cow files (Wenz and Giebel, 2012), and the EVENTS\5O FOR LACT = 0 command was used to obtain PWC files. Farms were visited only once; during a farm visit, animal health records were reviewed, and a survey was used to collect additional information from owners or farm workers who were responsible for animal care. Treatment data for postweaning heifers were not collected because many farmers sent these animals to other locations, and disease recording systems were not considered reliable for these age groups. Although few farms retained bull calves, antimicrobial treatment of PWC could have included bulls because AMU was quantified for all animals under 60 d of life. Farmers received a $100 incentive for their participation. All questions referred to the 1-yr period preceding the day of the farm visit.

Additional data were obtained using a survey instrument that contained 137 questions (available in supplemental materials). The survey was adapted from a previous study (Pol and Ruegg, 2007) and was administered by a single individual (J. L. C.) during the farm visit. Farm owners or herd-health managers were questioned about farm structure and demographics (17 questions), inventory (5 questions), replacement management (5 questions), antimicrobial treatment records for lactating cows (4 questions) and calves (2 questions), disease treatment or preventive practices in adult cows (80 questions) and calves (14 questions), veterinary feed directive (5 questions), veterinary involvement (3 questions), and drug purchase (2 question). To aid in the identification of antimicrobials and to confirm label information, laminated pages containing full color pictures of commercially available veterinary antimicrobial drugs for oral, systemic, and intramammary (**IMM**) use were shown to interviewees.

Estimation of Antimicrobial Usage

Antimicrobial usage was quantified using a standard unit referred to as ADD (Jensen et al., 2004), following a methodology described in previous studies (Pol and Ruegg, 2007; Saini et al., 2012; Mills et al., 2018). For each active ingredient, a standard ADD was calculated (Table 1) and defined as the maximum antimicrobial dose per day that an animal would receive using the label dosages approved by the Food and Drug Administration (**FDA**). Holsteins were the predominant breed, but farms contained some Brown Swiss, Jerseys, or crossbred animals (Table 2). As the average estimated BW of adult cows was 678 $\pm$ 9.9 kg among farms, a BW of 680 kg for adult cows (Pol and Ruegg, 2007) and a BW of 64 kg for PWC (Jones and Heinrichs, 2020) were used to estimate the standard ADD. Approved dosages for licensed animal drugs were obtained from the US National Library of Medicine–DailyMed (https://dailymed.nlm.nih.gov/dailymed/index.cfm), and dosages for antimicrobials not approved were estimated using dosages from reputable veterinary manuals (Aiello et al., 2016; for 1 antimicrobial, trimethoprim-sulfa, from personal communication with a faculty member at the University of Wisconsin-Madison School of Veterinary Medicine).

Animal defined doses for IMM AMU were calculated using the following formula:

$$\mathrm{ADD_{IMMA}} = \frac{\text{quarters treated} \times \left(\frac{\text{tubes}}{\text{application}}\right) \times \left(\frac{\text{applications}}{\text{day}}\right) \times \text{days}}{\mathrm{ADD_{IMMstandard}}},$$

where $\mathrm{ADD_{IMMA}}$ is the final ADD for intramammary antimicrobial A; quarters treated is the number of quarters treated with antimicrobial A; tubes per application is the number of tubes used per treatment; applications per day is the frequency that antimicrobial A is administered per day; days is the total number of days that antimicrobial A was administered; and $\mathrm{ADD_{IMMstandard}}$ is the standard ADD for antimicrobial A.

Animal defined doses for systemic and oral antimicrobial treatments, were calculated using the following formula:

$$\mathrm{ADD_{SYS/ORALA}} = \frac{\text{concentration} \times \text{maximal dose} \times \left(\frac{\text{applications}}{\text{day}}\right) \times \text{days}}{\mathrm{ADD_{SYS/ORALstandard}}},$$

Table 1. Animal daily doses (ADD) used for estimating antimicrobial usage in adult cows and preweaned calves on 40 Wisconsin dairy farms[1]

Active ingredient	Product type[2]	Route[3]	Concentration	Times per day	Dosage	ADD	
						Adult cows[4]	Preweaned calves[4]
Amoxicillin	RX	IMM	62.5 mg/tube	2	1 tube	2 tube (s)	—
Ceftiofur	RX	IMM	125 mg/tube	1	1 tube	1 tube (s)	—
Cephapirin	OTC	IMM	200 mg/tube	2	1 tube	2 tube (s)	—
Hetacillin	RX	IMM	62.5 mg/tube	1	1 tube	1 tube (s)	—
Pirlimycin	RX	IMM	50 mg/tube	1	1 tube	1 tube (s)	—
Cloxacillin	RX	IMM	200 mg/tube	2	1 tube	2 tube (s)	—
Ceftiofur	RX	IMM	500 mg/tube	1	1 tube	1 tube	—
Cephapirin	OTC	IMM	300 mg/tube	1	1 tube	1 tube	—
Cloxacillin	RX	IMM	500 mg/tube	1	1 tube	1 tube	—
Cloxacillin	RX	IMM	500 mg/tube	1	1 tube	1 tube	—
Novobiocin; penicillin G	OTC	IMM	400 mg/200,000 IU	1	1 tube	1 tube	—
Dihydrostreptomycin sulfate; pen G	RX	IMM	1,000 mg; 1,000,000 IU	1	1 tube	1 tube	—
Ampicillin	RX	IM	250 mg/mL	1	11 mg/kg	7,480 mg	704 mg
Ceftiofur	RX	SQ	200 mg/mL	1	6.6 mg/kg	4,488 mg	422.4 mg
Ceftiofur	RX	IM, SQ	50 mg/mL	1	2.2 mg/kg	1,496 mg	140.8 mg
Danofloxacin	RX	SQ	180 mg/mL	1	6 mg/kg	—	384 mg
Enrofloxacin (single dose)	RX	SQ	100 mg/mL	1	12.5 mg/kg	—	800 mg
Enrofloxacin (multiple doses)	RX	SQ	100 mg/mL	1	5 mg/kg	—	320 mg
Florfenicol (single dose)	RX	IM	300 mg/mL	1	40 mg/kg	—	2,560 mg
Florfenicol (multiple doses)	RX	SQ	300 mg/mL	1	20 mg/kg	13,600 mg	1,280 mg
Florfenicol	RX	SQ	300 mg/mL		40 mg/kg	—	2,560 mg
Gamithromycin	RX	SQ	150 mg/mL	1	6 mg/kg	—	384 mg
Gentamicin	RX	IM	100 mg/mL	1	12 mg/kg	—	768 mg
Oxytetracycline (single dose)	OTC	SQ	200 mg/mL	1	19.8 mg/kg	13,464 mg	1,267 mg
Oxytetracycline (multiple doses)	OTC	SQ, IV	200 mg/mL	1	11 mg/kg	7,480 mg	704 mg
Oxytetracycline (single dose)	OTC	IM, SQ	200 mg/mL	1	19.8 mg/kg	13,464 mg	1,267 mg
Oxytetracycline (multiple doses)	OTC	IM, IV, SQ	200 mg/mL	1	11 mg/kg	7,480 mg	704 mg
Oxytetracycline	OTC	IV	100 mg/mL	1	11 mg/kg	7,480 mg	—
Penicillin G procaine	OTC	IM	300,000 IU/mL	1	6,614 IU/kg	4,497,520 mg	423,296 mg
Spectinomycin	RX	SQ	100 mg/mL	1	15 mg/kg	10,200 mg	—
Sulfadimethoxine	OTC	IV	400 mg/mL	1	33 mg/kg	22,440 mg	2,112 mg
Tildipirosin	RX	SQ	180 mg/mL	1	4 mg/kg	—	256 mg
Tilmicosin	RX	SQ	300 mg/mL	1	10 mg/kg	6,800 mg	640 mg
Tulathromycin	RX	SQ	100 mg/mL	1	2.5 mg/kg	1,700 mg	160 mg
Tylosin	OTC	IM	200 mg/mL	1	17.6 mg/kg	11,968 mg	1,126.4 mg
Lincomycin; spectinomycin	RX	IM	—	—	10 mg/kg	6,800 mg	—
Sulfadimethoxine	OTC	ORAL	5,000; 15,000 mg/bolus	1	33.1 mg/kg	22,508 mg	2,118.4 mg
Trimethoprim-sulfamethoxazole	RX	ORAL	960 mg/bolus	1	20 mg/kg	—	1,280 mg
Oxytetracycline	OTC	ORAL	500 mg/bolus	2	22 mg/kg	—	1,408 mg

[1]Data were obtained using the current label listed in the US National Library of Medicine, veterinary manuals (gentamicin and lincomycin-spectinomycin; Aiello et al., 2016), or veterinary school recommendations of Wisconsin sick calf protocols (trimethoprim-sulfamethoxazole; McGuirk, 2018).

[2]RX = prescription animal drug; OTC = over the counter.

[3]IMM = intramammary infusion; IM = intramuscular; IV = intravenous; SQ = subcutaneous.

[4]A BW of 680 kg was used for lactating dairy cows and 64 kg for preweaned calves.

where $ADD_{SYS\backslash ORALA}$ is the final systemic or oral ADD for antimicrobial A; concentration is the milligram or international unit per milliliter of antimicrobial A; maximal dose is the maximal dosage of antimicrobial A; applications per day is the frequency that antimicrobial A was administered per day; days is the total number of days that antimicrobial A was administered; and $ADD_{SYS/ORALstandard}$ is the standard ADD for antimicrobial A.

Similar to Saini et al. (2012) and Grave et al. (1999), the ADD of trimethoprim-sulfamethoxazole combination was based on the active ingredient (trimethoprim) dosage. Similar to Saini et al., (2012), international unit of the IMM combination compounds (novobiocin sodium and penicillin G procaine; dihydrostreptomycin sulfate; penicillin G procaine) were converted to milligrams using a conversion of 1,000 IU of penicillin G procaine equal to 0.6 mg of penicillin G procaine. When the labeled dose included a range, an average dose was calculated using the initial and subsequent dose values to calculate an average dose according to the maximum treatment days described on label. Each dry cow

Table 2. Characteristics of Wisconsin dairy herds (n = 40) enrolled in a study about antimicrobial usage in September to December, 2017

Farm	RHA[1] (kg/cow per year)	BTCC[2] (cells/mL)	Adult cows[3] (n)	Preweaned calves[4] (n)	Parity (%)			Adult cows BW[5] (kg)	Holstein cows[6] (%)
					First	Second	≥ Third		
1	13,141	122,000	2,781	337	40	29	30	726	100
2	14,878	80,000	469	65	44	28	28	721	95
3	13,313	175,000	598	65	34	27	39	499	100
4	13,608	110,000	1,559	201	38	30	33	656	99
5	14,389	78,000	332	49	41	30	30	726	100
6	13,103	120,000	816	92	38	27	34	634	99
7	12,928	86,000	393	53	37	28	35	590	100
8	13,140	187,000	2,357	314	37	30	33	679	97
9	10,829	195,000	2,382	320	45	30	25	724	99
10	14,061	105,000	623	95	46	30	25	726	100
11	12,505	181,000	1,197	178	35	30	35	680	100
12	13,298	121,000	475	64	35	30	35	612	99
13	14,075	188,000	2,152	283	47	30	23	703	100
14	13,381	126,000	454	46	38	31	31	725	99
15	14,334	155,000	734	—	44	23	33	787	90
16	14,375	146,000	604	97	37	26	37	612	100
17	12,993	118,000	2,031	333	43	32	24	680	100
18	13,608	140,000	1,016	130	35	28	37	635	100
19	14,742	162,000	762	99	38	27	35	649	95
20	14,061	115,000	583	67	39	29	32	590	100
21	13,154	120,000	3,070	—	35	27	38	634	99
22	10,905	128,000	5,005	861	40	30	30	741	98
23	10,925	256,000	1,615	—	37	29	34	791	99
24	13,337	142,000	1,160	182	36	29	34	631	77
25	12,353	133,000	887	107	32	35	33	647	86
26	13,117	150,000	676	109	39	30	31	629	96
27	13,517	105,000	581	86	39	30	31	658	100
28	13,154	110,000	592	50	40	21	39	702	100
29	12,775	60,000	441	37	45	29	26	629	88
30	15,059	174,000	586	70	41	25	34	771	100
31	13,517	320,000	954	126	41	31	28	748	99
32	14,288	160,000	598	64	0	0	0	680	100
33	14,601	188,000	443	49	37	24	39	658	100
34	13,608	164,000	1,415	184	39	27	35	680	100
35	13,081	165,000	954	160	34	37	28	680	99
36	13,381	148,000	1,448	262	45	34	21	701	98
37	13,245	77,000	3,444	—	36	36	28	771	97
38	13,608	129,000	1,189	157	38	25	38	771	100
39	12,530	137,000	1,527	—	46	27	26	622	99
40	10,886	126,000	3,736	889	41	33	26	633	98
Mean	13,295	142,600	1.316	180	38	28	31	678	99

[1]Rolling herd average.

[2]Bulk tank SCC from the month preceding farm visit.

[3]Total lactating and dry cows.

[4]<60 d.

[5]Estimated BW.

[6]Weighted by proportion of predominant breeds.

therapy (**DCT**) tube was defined as 1 ADD, resulting in a total of 4 ADD for most cows administered DCT (Scherpenzeel et al., 2014; Stevens et al., 2016). Due to challenges in adequately measuring AMU in foot baths, wound sprays, and intraocular sprays, these antimicrobials were not included in the analysis. Although we collected information about usage of ionophores, we did not include them in analysis as they are not considered medically important.

Dose-based estimates (ADD) were estimated at herd level, for cows, and for PWC. Herd-level AMU per animal was defined as the sum of the ADD used on a farm divided by the total animals at risk during the 365-d period [(adult cows + PWC) × 365 d × 1,000]. For cows, AMU was defined as the sum of the ADD used in adult cows divided by the average adult cows at risk during the 365-d period (average adult cows × 365 d × 1,000). Dose-based AMU in PWC was estimated as sum of the ADD used in PWC divided by the average PWC at risk during the 365-d period (PWC × 365 d × 1,000). Animals at risk were estimated based on number of adult cows or PWC as indicated in the dairy management software during the month of visit.

Mass-based estimates (total mg of antimicrobial per kg of animal weight) were calculated following methodology as described by Mills et al. (2018). In brief, total milligrams of antimicrobials for IMM compounds were calculated using the following formula:

$$\text{Total}_{\text{IMMA}} = \frac{\text{quarters treated} \times \left(\frac{\text{applications}}{\text{day}}\right) \times \text{days} \times \text{concentration}}{\text{animals at risk} \times \text{BW}},$$

where $\text{Total}_{\text{IMMA}}$ (mg/kg) is the final milligrams for intramammary antimicrobial A per kilogram of BW; quarters treated is the number of quarters treated with antimicrobial A; applications per day is the frequency that antimicrobial A was administered per day; days is the total number of days that antimicrobial A was administered; concentration is the milligram per milliliter of antimicrobial A; and animals at risk × BW is the number of animals at risk times the standard BW defined for each animal class.

Total milligrams of antimicrobials for systemic and oral compounds were calculated using the following formula:

$$\text{Total}_{\text{SYS/ORALA}} = \frac{\text{concentration} \times \text{maximal dose} \times \left(\frac{\text{applications}}{\text{day}}\right) \times \text{days}}{\text{animals at risk} \times \text{BW}},$$

where $\text{Total}_{\text{SYS/ORALA}}$ (mg/kg) is the final milligrams for systemic or oral antimicrobial A per kilogram of BW; concentration is the milligram per milliliter of antimicrobial A; maximal dose is the maximal dosage of antimicrobial A; applications per day is the frequency that antimicrobial A was administered per day; days is the total number of days that antimicrobial A was administered; and animals at risk × BW is the number of animals at risk times the standard BW defined for each animal class.

Mass-based calculations were estimated at the herd level, for cows, and for PWC. For cows, AMU was defined as the sum of the milligrams of antimicrobial used divided by the number of adult cows multiplied by a standard BW. Antimicrobial usage in PWC was defined as the sum of the milligrams of antimicrobial used divided by the number of PWC multiplied by a standard BW. A standard BW of 680 kg was used for lactating cows (Pol and Ruegg, 2007) and 64 kg for PWC (Jones and Heinrichs, 2020). At the herd level, AMU was defined as the sum of the total milligrams of antimicrobial used divided by animal BW (cow weight + PWC weight).

Statistical Analysis

Statistical analyses were performed using SAS version 9.4 (SAS Institute, Cary, NC). Descriptive statistics were performed using PROC MEANS and used to characterize participating herds and summarize AMU by active ingredient, route, and animal class. Analysis of variance was performed using GLIMMIX with dependent variable of ADD or Total (mg/kg of BW), and independent variables were active ingredients or route of administration as follows:

$$Y_i = \mu + \tau_i + e_i,$$

where Y_i = the dependent variable, $\mu + \tau_i$ is the effect of active ingredient or route, and e = the residual error. Farm was the experimental unit. Route of usage (oral, injectable, IMM) and active ingredient (Table 1) were defined as categorical variables, and values for ADD and mass-based estimates were defined as continuous variables. Normality of the data was evaluated using normal probability and box plots with PROC UNIVARIATE, and normality of residuals was evaluated based on plots of residuals versus predicted values. A natural log-transformation was used for ADD and mass estimates to normalize the distributions

Statistical analyses were performed only for antimicrobials used on ≥5 herds. The null hypothesis was that AMU did not vary by route or active ingredient, and statistical difference was considered when $P < 0.05$.

RESULTS

Characteristics of Herds

Of farms that received invitation letters (n = 413), 109 (26%) responded, with 80 affirmative and 29 negative responses. After phone interviews with affirmative responders, 43 farmers remained interested in participating, whereas 16 farmers could not be contacted or were not eligible, and 21 declined participation. From the 43 eligible farms, a convenience sample of 40 herds was enrolled based on ease of scheduling and the need to identify qualifying herds for enrollment in a broader study that includes additional objectives related to AMR.

Enrolled farms were distributed across Wisconsin. Based on cow numbers found in dairy management records collected at enrollment, enrolled farms contained 52,639 (mean = 1,316 ± 169) adult cows and 6,281 PWC (mean = 180 ± 33; Table 2). Cows were housed in freestalls containing fresh sand (n = 22), recycled sand (n = 9), manure solids (n = 4), wood products (n = 2), or mixed bedding materials (n = 3). The rolling herd average was 13,295 ± 164.4 kg of milk/cow per year and ranged from 10,829 to 15,059 kg of milk/cow per year. Average bulk tank SCC was 143,600 ± 7,600 cells/mL and ranged from 60,000 to 320,000 cells/mL. Among parity groups, the greatest proportion of adult animals was in first lactation (Table 2). Of enrolled farms, 35 raised PWC on the farm, and 5 sent PWC to other specialized locations.

Treatment Records Obtained from Computerized Records

Enrolled farms included 96,431 treatment remarks, in which 76,239 remarks were related to treatments given to adult cows, and the remaining 20,192 to treatments given to PWC. Of total treatment remarks for adult cows, 78% (59,213) were for IMM treatments (36,161 remarks for DCT, 23,042 remarks for treatment of lactating cows, 10 remarks for intraocular treatment using IMM product), 22% (16,913) were for systemic treatment, and the remaining 0.2% (113) were for oral treatments. When events from dairy management software included a treatment protocol that did not specify the actual number of days for which the treatment was administered, treatment days were obtained from survey data for that farm. Records for PWC contained 20,192 remarks, of which 90% (18,100) were for systemic treatments, and the remaining 10% (2,092) were for oral treatments.

Of the total remarks related to adult cows, about 99% (76,171) were extracted from computerized records, with approximately 94% (71,839) of adult cow health records obtained from electronic records. The remaining 6% (4,332) were also obtained from electronic records, but had to be manually entered because researchers did not have access to the software used by 3 farms. Of farms using IMM antimicrobials to treat lactating cows (n = 39), almost every farm used a dairy management program as their only mechanism for recording IMM antimicrobial treatment of lactating dairy cows, with the exception of 2 farms that also kept paper files of treatments, such that only 36 IMM treatment events had to be manually entered. All IMM dry-off treatments and records of oral treatments were obtained from dairy management software, and less than 1% (32 treatments) of systemic treatments were obtained from paper records or based on recall. Approximately 87% (17,584) of PWC animal health records were acquired from computerized records, with 16,277 (93%) remarks obtained from electronic records. The remaining 1,307 (7%) remarks were also obtained from electronic records, but had to be manually entered because researchers did not have access to the software used by 3 farms. Of the remaining 13% (2,608) of the animal health records, about 6% (159 treatments) were oral records, and 94% (2,449) of the systemic treatments were obtained from paper records or based on recall and had to be entered manually.

Overall AMU in Adult Cows and PWC

In accordance with enrollment criteria, all farms reported AMU for treatment or prevention in adult cows, and 35 farms reported AMU in PWC. At herd level and across animal classes, 24 active ingredients were reported. Ceftiofur was the only antimicrobial used on all farms, followed by ampicillin (n = 36) and tulathromycin (n = 30). Three antimicrobials were used only on a single farm (Table 3).

For cows across all routes of administration, 18 different active ingredients were used for treatment of adult dairy cows (Table 4). Ceftiofur and cephapirin were most commonly used, and all farms reported use of ceftiofur. Ceftiofur was administered as an IMM preparation at dry off (23 farms), for treatment of mastitis during lactation (36 farms), or as an injectable product (40 farms). Of all antimicrobial treatments given to adult cows, 5 products were used by just 1 farm. Four products used in lactating cows (florfenicol, tylosin, lincomyin, and lincomycin-spectinomycin combination) were not approved for use in this class of animals, but can be used under guidance of a veterinarian based on extralabel usage guidelines. Two products given to adult cows (tulathromycin and tilmicosin) are not labeled for use in that class of animals, and FDA regulations en-

force zero tolerance for residues. Therefore, extremely long milk withholding periods are recommended if these products are used (FARAD, 2021). All farms reported administration of antimicrobials using both IMM and systemic routes, and 4 reported administration of oral antimicrobials (Table 5). Use of IMM antimicrobials at dry off was reported by all farms, and use of IMM antimicrobials for treatment of lactating cows was reported by 39 farms. Intramammary tubes were used for intraocular treatment of a few cases of infectious bovine keratoconjunctivitis (pink eye) in adult cows on 1 farm.

Fifteen different active ingredients were used for treatment of PWC (Table 6). All farms that had PWC on site reported usage of injectable antimicrobials for treatment of PWC, and 8 farms reported usage of antimicrobials administered orally (Table 5). Ceftiofur, enrofloxacin, florfenicol, penicillin G, and tulathromycin were reported to be used by at least half of the farms (Table 6). All antimicrobials given to PWC were either approved for use in this class of animal or permitted for use under extralabel usage guidelines. Use of ionophores was reported by 38 farms, with 11 reporting usage for both adult cows and PWC, 25 only for adult cows, and 2 farms for growing heifers.

Quantification of AMU Using a Dose-Based Metric

By Farm and Animal Class. Mean herd-level ADD (denominator included adult cows and PWC) was 17.2 ± 1.1 ADD/1,000 animal-d per farm and ranged from 6.1 to 42.6 ADD/1,000 animal-d (Figure 1A). For all 40 enrolled farms, combined herd-level ADD totaled 687.6 ADD/1,000 animal-d. Proportionally, ceftiofur (46%), cephapirin (13%), penicillin G (9%), ampicillin (6%), and dihydrostreptomycin-penicillin (5%) totaled 79% of total herd-level ADD per 1,000 animal-d (Table 3). The remaining 21% of ADD was contributed by 19 antimicrobials. Antimicrobial usage varied among active ingredients ($P < 0.001$; Table 3). For active ingredients used on > 5 farms, herd-level ADD (back transformed LSM) were greatest for ceftiofur, cephapirin, dihydrostreptomycin, and cloxacillin (Table 3), and there was a tendency for greater herd-level ADD for ceftiofur as compared with cephapirin (5.33 vs. 1.22

Table 3. Total antimicrobial usage administered to preweaned calves and adult cows on 40 Wisconsin dairy farms for a 1-yr period estimated using animal daily doses (ADD; per 1,000 animal-d) or mass density (total milligrams of antimicrobial per kilogram of animal weight)

		ADD per 1,000 animal-days					Mass-based (mg/kg of drug per BW)				
Active ingredient[1]	Farm (n)	LSM[2,3]	SEM	Median	Total	(%)	LSM[2,3]	SEM	Median	Total	(%)
Amoxicillin	6	0.17^{bcd}	2.05	0.18	2.89	0.42	0.02^{cdef}	2.07	0.03	0.43	0.08
Ampicillin	36	0.62^{bcd}	1.35	0.72	43.46	6.32	2.07^{a}	1.35	3.12	161.41	29.60
Ceftiofur	40	5.33^{a}	1.33	6.56	315.39	45.86	2.21^{a}	1.33	2.34	140.68	25.80
Cephapirin	24	1.22^{abc}	1.44	1.62	86.85	12.63	0.35^{abc}	1.44	0.63	19.11	3.51
Cloxacillin	8	1.57^{abc}	1.87	4.80	31.18	4.53	0.44^{abc}	1.88	1.38	9.15	1.68
Danofloxacin	1	—	—	0.07	0.07	0.01	—	—	0.01	0.01	<0.01
Dihydro-streptomycin	9	2.76^{ab}	1.80	3.86	36.14	5.26	2.73^{a}	1.82	3.75	34.87	6.40
Enrofloxacin	27	0.16^{cd}	1.41	0.21	12.01	1.75	0.05^{cde}	1.42	0.06	3.59	0.66
Florfenicol	21	0.06^{d}	1.47	0.06	4.90	0.71	0.09^{dc}	1.48	0.09	9.46	1.74
Gamithromycin	11	0.05^{d}	1.70	0.10	1.89	0.27	0.01^{def}	1.72	0.02	0.49	0.09
Gentamicin	2	—	—	0.01	0.02	<0.01	—	—	0.01	0.01	<0.01
Hetacillin	12	0.38^{bcd}	1.66	0.47	7.74	1.13	0.01^{def}	1.68	0.02	0.29	0.05
Linco-spectinomycin[4]	1	—	—	0.14	0.14	0.02	—	—	0.02	0.02	<0.01
Oxytetracycline	25	0.10^{d}	1.43	0.09	9.54	1.39	0.33^{abc}	1.43	0.40	32.94	6.04
Penicillin G	25	0.59^{bcd}	1.43	0.88	60.45	8.79	0.20^{bcd}	1.43	0.30	35.26	6.47
Penicillin novobiocin	6	0.76^{abcd}	2.05	2.37	14.27	2.08	0.24^{abcd}	2.07	0.74	4.52	0.83
Pirlimycin	19	0.17^{cd}	1.50	0.18	19.16	2.79	0.01	1.51	<0.01	0.57	0.10
Spectinomycin	1	—	—	0.04	0.04	0.01	—	—	0.22	0.22	0.04
Sulfadimethoxine	13	0.12^{cd}	1.63	0.11	15.39	2.24	1.15^{ab}	1.64	1.42	65.80	12.07
Tildipirosin	4	—	—	0.02	0.09	0.01	—	—	<0.01	0.01	<0.01
Tilmicosin	5	—	—	0.05	0.32	0.05	—	—	0.06	0.81	0.15
TMP-sulfa[5]	5	—	—	3.24	13.77	2.00	—	—	4.99	21.80	4.00
Tulathromycin	30	0.09^{d}	1.38	0.08	11.65	1.69	0.01^{ef}	1.39	0.01	1.67	0.31
Tylosin	2			0.15	0.29	0.04	—	—	1.05	2.10	0.39

[a–f]Mean values within the same column with different superscripts differ from each other ($P < 0.001$).

[1]Active ingredients used on farm in either adult cows or preweaned calves or both.

[2]Active ingredients used on ≤5 farms were not used in analysis among means.

[3]Statistical analyses were performed on natural logs; data are presented as back transformed LSM.

[4]Lincomycin-spectinomycin combination.

[5]Trimethoprim (TMP)-sulfamethoxazole combination.

Table 4. Comparison between dose-based (animal daily doses per 1,000 cow-d) or mass-based (total milligrams of antimicrobial per adult cow BW in kilograms) metrics used to estimate antimicrobial usage on 40 WI dairy farms for a 1-yr period

Active ingredient	Farm (n)	Animal daily dose per 1,000 cow-days					Mass-based (mg/kg of drug per BW)				
		LSM[1,2]	SEM	Median	Total	(%)[3]	LSM[1,2]	SEM	Median	Total	(%)[3]
Intramammary dry cow product											
Ceftiofur	23	3.26	1.37	7.36	131.55	42.49	0.89	1.36	1.98	35.31	35.94
Cephapirin	14	4.51	1.50	7.54	86.04	27.79	0.95	1.51	1.23	13.81	14.05
Cloxacillin	7	3.05	1.78	6.81	34.43	11.12	0.81	1.75	1.83	9.24	9.40
Penicillin dihydrostreptomycin	9	2.79	1.66	4.43	41.13	13.29	2.41	1.64	3.81	35.32	35.95
Penicillin novobiocin	6	0.95	1.86	2.67	16.42	5.30	0.27	1.83	0.75	4.58	4.66
Intramammary lactating cow product											
Amoxicillin	6	0.18[b]	1.93	0.20	3.23	1.75	0.02[ab]	1.93	0.03	0.43	2.68
Ceftiofur	36	2.32[a]	1.31	3.02	137.79	74.83	0.16[a]	1.31	0.20	9.24	57.57
Cephapirin	17	0.30[b]	1.48	0.49	12.78	6.94	0.13[a]	1.48	0.21	5.49	34.21
Cloxacillin	1	—	—	0.01	0.01	0.01	—	—	0.01	0.01	0.06
Hetacillin	12	0.40[b]	1.59	0.55	8.80	4.78	0.01[b]	1.59	0.02	0.30	1.87
Pirlimycin	19	0.19[b]	1.45	0.18	21.52	11.69	0.01[b]	1.45	<0.01	0.58	3.61
Oral product											
Sulfadimethoxine	4	—	—	0.44	2.27	100.00	—	—	5.34	27.45	100.00
Injectable products											
Ampicillin	34	0.63[a]	1.31	1.29	39.82	28.99	2.49[a]	1.31	3.26	159.89	46.55
Ceftiofur	40	0.85[a]	1.28	2.36	68.76	50.05	1.12[ab]	1.29	1.04	96.37	28.06
Florfenicol	1	—	—	0.43	0.43	0.31	—	—	3.16	3.16	0.92
Lincomycin-spectinomycin[4]	1	—	—	0.15	0.15	0.11	—	—	0.02	0.02	0.01
Oxytetracycline	23	0.07[bc]	1.38	0.31	6.17	4.49	0.33[c]	1.39	0.32	29.89	8.70
Penicillin G	14	0.24[ab]	1.51	1.47	19.73	14.36	0.34[bc]	1.51	0.40	28.81	8.39
Spectinomycin	1	—	—	0.04	0.04	0.03	—	—	0.22	0.22	0.06
Sulfadimethoxine	9	0.04[c]	1.67	0.26	1.86	1.35	0.40[bc]	1.67	0.70	22.36	6.51
Tilmicosin	2	—	—	0.07	0.09	0.07	—	—	0.31	0.63	0.18
Tulathromycin	1	—	—	<0.01	<0.01	<0.01	—	—	0.01	0.01	<0.01
Tylosin	1	—	—	0.33	0.33	0.24	—	—	2.12	2.12	0.62

[a,b]Mean values within the same product type and column with different superscripts differ from each other ($P < 0.001$).

[1]Active ingredients containing 5 or less farms were not used in the comparison among means.

[2]Statistical analyses were performed using natural logs; data are presented as back transformed LSM.

[3]Proportion within route of antimicrobial usage.

[4]Lincomycin-spectinomycin combination.

Table 5. Estimated antimicrobial usage in adult cows and preweaned calves (PWC) on 40 Wisconsin dairy farms by route of administration for a 1-yr period using dose-based (animal daily doses; ADD) and mass-based (total mg/kg of antimicrobial per animal weight)

Route	Farms (n)	Animal daily dose (ADD/1,000 cow-d; ADD per 1,000 PWC-d)					Mass-based (mg/kg of drug per BW)				
		LSM[1,2]	SEM	Median	Total	(%)[3]	LSM[1,2]	SEM	Median	Total	(%)[3]
Adult cow											
Injectable	40	2.22[b]	1.13	2.48	137.38	21.69	5.12[a]	1.15	5.79	343.47	70.78
Intramammary[4]	40	11.69[a]	1.13	11.40	493.72	77.95	2.56[b]	1.15	2.32	114.37	23.56
Oral	4	—	—	0.44	2.27	0.36	—	—	5.34	27.45	5.66
Preweaned calf											
Injectable	35	11.22	1.30	19.40	783.74	79.10	5.39	3.60	38.85	2,139.42	41.84
Oral	8	9.22	1.72	17.00	207.11	20.90	148.32	14.55	321.37	2,973.74	58.16

[a,b]Mean values within the same column with different superscripts differ from each other ($P < 0.001$).

[1]Differences among routes were not estimated if ≤5 farms reported use of compound.

[2]Statistical analyses were performed using natural logs; data are presented are back transformed LSM.

[3]Proportion within class of animal.

[4]Intramammary animal daily doses include antimicrobials used in mastitis treatment, dry cow therapy, and intraocular treatment.

ADD/1,000 animal-d; $P = 0.07$). Among farms that contained both PWC and adult cows (n = 35), AMU in adult cows represented 83% (95% CI: 78, 88) of the combined herd-level ADD and ranged from 31 to 99.9% of the total AMU.

Mean ADD for cows (denominator is adult cows) was 15.8 ± 0.9 ADD/1,000 cow-d per farm and ranged from 6.1 to 29.8 ADD/1,000 cow-d. The combined ADD for cows in the entire 40 herds was 633.4 ADD/1,000 cow-d. Among antimicrobials, ceftiofur represented 53% of all ADD administered in adult cows. The second antimicrobial with the greatest proportion of ADD was cephapirin, representing 16% of all ADD given to adult cows.

Mean ADD for PWC (denominator is PWC) was 28.3 ± 5.4 ADD/1,000 PWC-d per farm and ranged from 0.3 to 135.4 ADD/1,000 PWC-d. The ADD for PWC on all 35 farms that contained PWC totaled 990.9 ADD/1,000 PWC-d per farm. For PWC, penicillin G was the antimicrobial that accounted for the greatest proportion of AMU, representing 32% of all ADD in PWC, and proportions of ceftiofur, enrofloxacin, sulfadimethoxine, trimethoprim-sulfa, and tulathromycin varied from 9 to 11% of ADD administered to PWC (Table 6).

Adult Cows—By Route of Administration. Intramammary, systemic, and oral routes were used to administer antimicrobials to adult cows (Table 5).

Table 6. Comparison of antimicrobial usage in preweaned calves by active ingredient estimated using dose-based (animal daily doses per 1,000 preweaned calf-d) and mass-based (total milligrams of antimicrobial per kilogram of preweaned calf BW) metric on 35 Wisconsin dairy farms

Active ingredient	Farm (n)	Animal daily dose per 1,000 preweaned calf-d					Mass-based (mg/kg of drug per BW)				
		LSM[1,2]	SEM	Median	Total	(%)	LSM[1,2]	SEM	Median	Total	(%)
Ampicillin	16	1.32[ab]	1.61	2.46	67.11	6.77	5.24[a]	1.61	9.89	269.46	5.27
Ceftiofur	20	0.56[b]	1.54	0.52	90.95	9.18	0.65[b]	1.53	0.53	83.21	1.63
Danofloxacin	1	—	—	0.86	0.86	0.09	—	—	1.89	1.89	0.04
Enrofloxacin	27	1.34[ab]	1.45	1.74	106.24	10.72	3.93[a]	1.45	4.70	270.12	5.28
Florfenicol	21	0.51[b]	1.52	0.53	42.96	4.34	6.82[a]	1.52	6.96	576.17	11.27
Gamithromycin	11	0.44[b]	1.77	0.71	14.97	1.51	1.07[ab]	1.76	1.68	35.27	0.69
Gentamicin	2	—	—	0.09	0.17	0.02	—	—	0.38	0.77	0.02
Oxytetracycline	11	0.60[ab]	1.77	2.17	44.07	4.45	3.85[ab]	1.76	11.83	365.68	7.15
Penicillin G	24	3.75[a]	1.49	3.09	315.32	31.82	5.53[a]	1.48	4.52	460.36	9.00
Sulfadimethoxine	3	—	—	9.30	102.86	10.38	—	—	292.12	1,422.35	27.82
Tildipirosin	4	—	—	0.20	0.99	0.10	—	—	0.29	1.45	0.03
Tilmicosin	5	—	—	0.41	2.16	0.22	—	—	2.98	15.79	0.31
TMP-sulfa[3]	5	—	—	24.01	100.06	10.10	—	—	350.62	1,460.88	28.57
Tulathromycin	30	0.80[ab]	1.43	0.70	102.05	10.30	0.79[b]	1.42	0.67	149.44	2.92
Tylosin	1	—	—	0.05	0.05	0.01	—	—	0.33	0.33	0.01

[a,b]Mean values within the same column with different superscripts differ from each other ($P < 0.05$).

[1]Active ingredients containing 5 or less farms were not used in the comparison among means.

[2]Statistical analyses were performed on natural logs for farms using the active ingredient; data are presented are back transformed LSM.

[3]Trimethoprim (TMP)-sulfamethoxazole combination.

When estimated using ADD, IMM administration was the primary route that antimicrobials were administered (Table 5). For adult cows, ADD of IMM products were almost 5 times greater than ADD of antimicrobials administered systemically ($P < 0.001$; Table 5) and represented 78% of total ADD given to adult cows. Intramammary treatment included antimicrobials given at dry off (63% of IMM ADD in adult cows) and for treatment of lactating cows (37% of IMM ADD in adult cows).

Mean ADD used for IMM treatment of lactating cows was 4.7 ± 0.6 ADD/1,000 cow-d per farm (Figure 2A) and ranged from 0.2 to 14.6 ADD/1,000 cow-d. Total ADD used for IMM treatment of lactating cows in herds that used IMM antimicrobials (n = 39) summed to 184.1/1,000 cow-d. Among IMM antimicrobials used in lactating cows, ceftiofur (75% of ADD) and cephapirin (7% of ADD) accounted for almost 82% of ADD.

All farms reported use of IMM antimicrobials for drying off cows. Mean ADD used for IMM treatment at dry off was 7.7 ± 0.2 ADD/1,000 cow-d per farm (Figure 2A) and ranged from 3.5 to 11.3 ADD/1,000 cow-d. Total ADD used for IMM treatment at dry off totaled 309.6 ADD/1,000 cow-d. Three β-lactam antimicrobials and 2 combination products were given at dry off (Table 4), but no differences in least squares means (**LSM**) ADD were observed among active ingredients ($P = 0.38$). Cephalosporin antimicrobials represented about 70% of ADD used at dry off (Table 4).

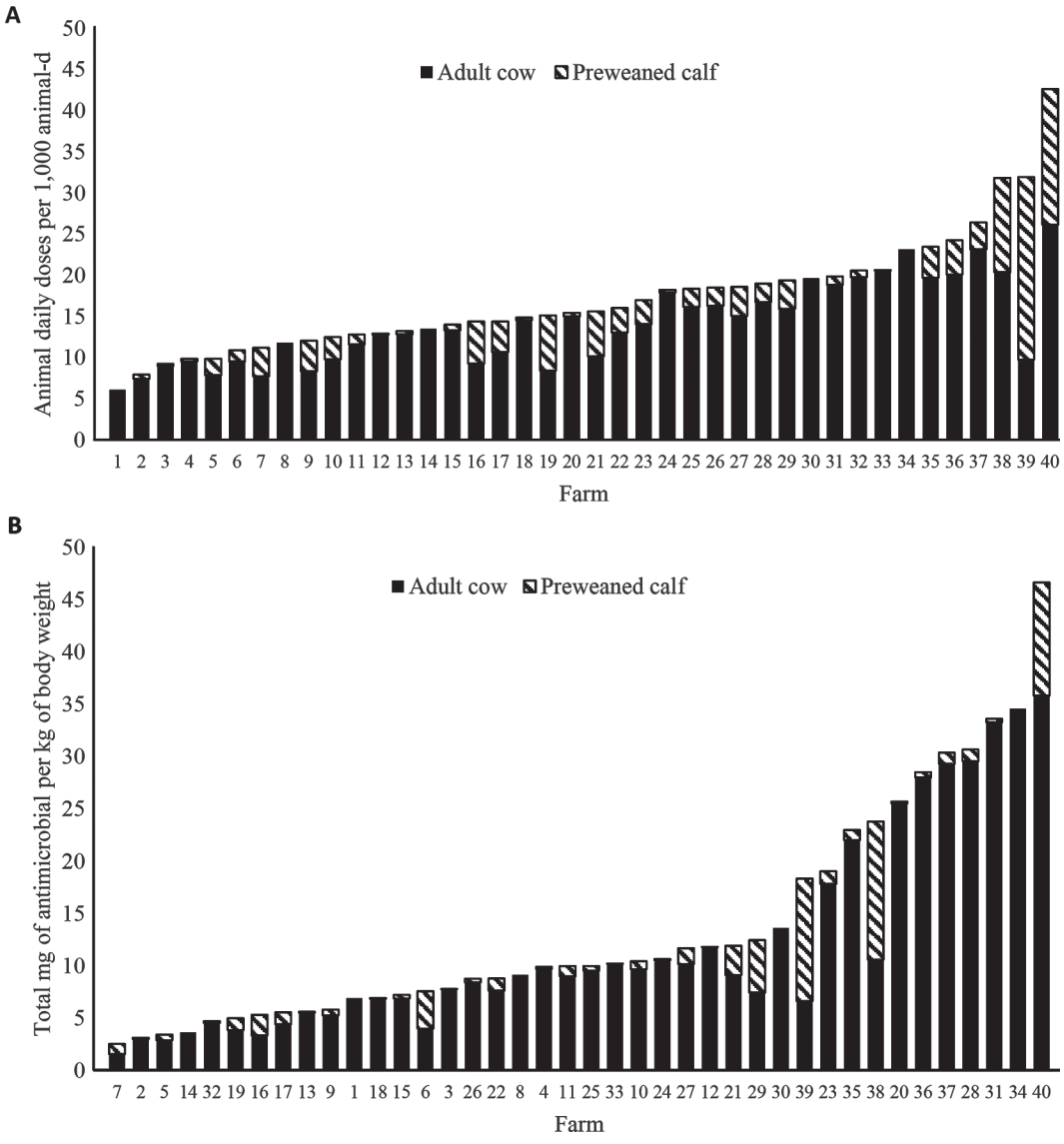

Figure 1. Herd-level antimicrobial usage for 40 Wisconsin dairy farms by animal category estimated using dose-based (A) and mass-based (B) metrics.

All farms reported use of injectable antimicrobials in adult cows (Table 5). Mean ADD administered systemically to adult cows were 3.4 ± 0.5 ADD/1,000 cow-d per farm (Figure 2A) and ranged from 0.1 to 14.1 ADD/1,000 cow-d. Total ADD used for systemic treatment of adult cows on all farms totaled 137.4 ADD/1,000 cow-d. Of 11 antimicrobials used systemically, 98% were accounted for by ceftiofur, ampicillin, penicillin G, and oxytetracycline (Table 4). Among active ingredients, ceftiofur accounted for most ADD (Table 4), but LSM ADD did not vary among ceftiofur, ampicillin, and penicillin G ($P = 0.89$; Table 4). Only 4 herds reported oral administration of antimicrobials in adult cows (Table 5). For adult cows, 0.6 ± 0.3 ADD

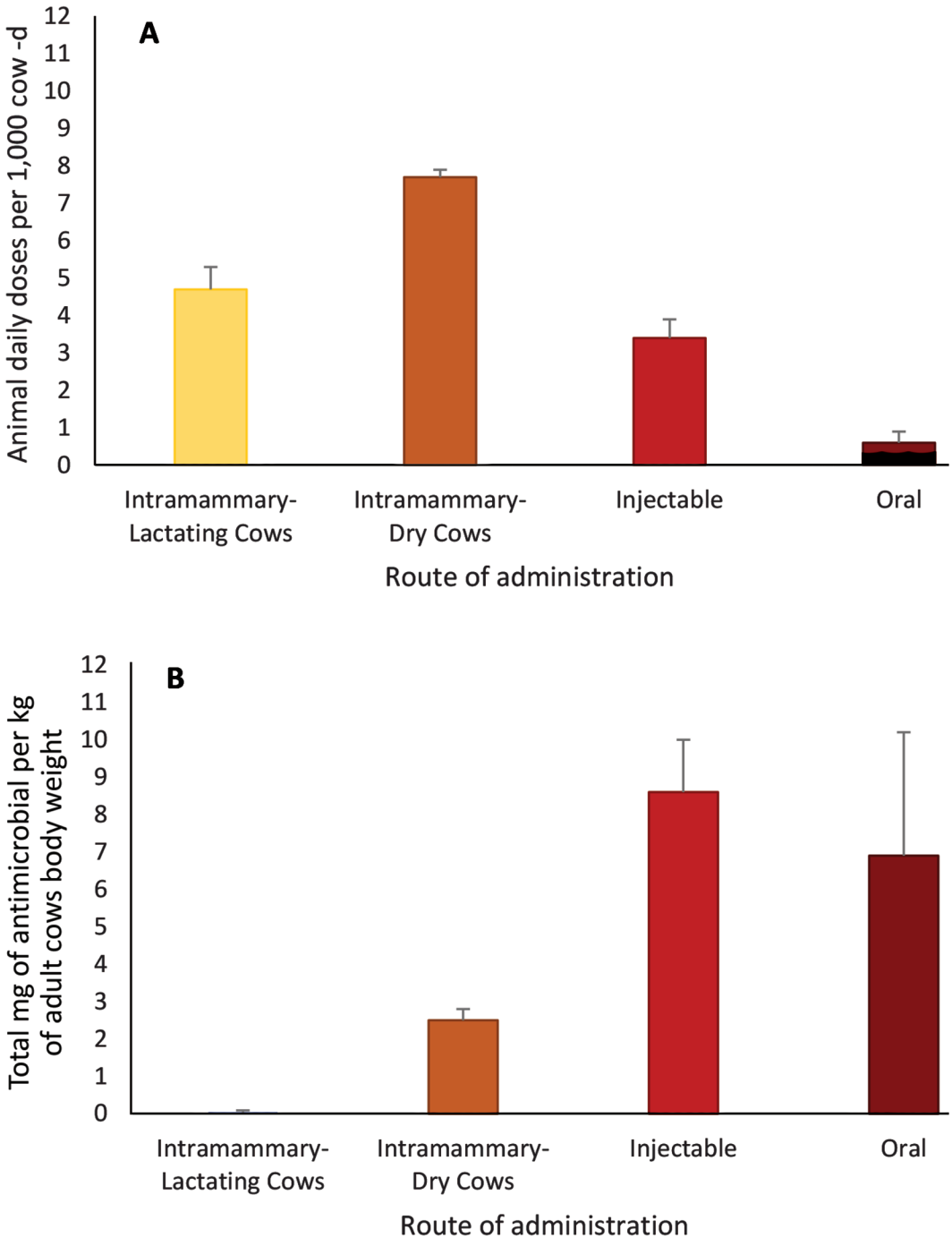

Figure 2. Antimicrobial usage in adult cows by route estimated using dose-based (A) and mass-based (B) metrics. Bars represent SEM.

/1,000 cow-d per farm were administered orally (Figure 2A). Total ADD given orally totaled 2.3 ADD/1,000 cow-d (Table 5).

PWC—By Route of Administration. Both injectable and oral antimicrobials were used in calves, but the number of ADD did not vary by route (Table 5; $P = 0.75$). For PWC, 22.4 ± 3.9 ADD/1,000 PWC-d per farm were given using an injectable route (Figure 3A) and ranged from 0.3 to 91.3 ADD/1,000 PWC-d. The combined ADD given to PWC using injections on all 35 farms that contained PWC summed to 783.7 ADD/1,000 PWC-d and represented 79% of all ADD given to PWC. For active ingredients used in greater than 5 herds using an injectable route, the adjusted mean ADD varied on active ingredient (Table 7; $P = 0.003$). Gamithromycin accounted for the fewest ADD, and penicillin G accounted for the greatest (Table 7). No difference in mean ADD was observed among ampicillin, enrofloxacin, and oxytetracycline (Table 7; $P = 0.64$).

For PWC, 25.9 ± 10.5 ADD/1,000 PWC-d were given orally (Figure 3A). The combined ADD of all antimicrobials given orally to PWC summed to 207.1 ADD/1,000 PWC-d and represented the remaining 21% of all AMU used in PWC. Among active ingredients given orally, the proportion of ADD were 5%, 47% and 48%, for oxytetracycline, sulfadimethoxine and trimethoprim-sulfamethoxazole combination, respectively (Table 7).

Quantification of AMU Using a Mass-Based Metric

By Farm and Animal Class. Mean herd-level antimicrobial mass density was 13.6 ± 1.7 mg of antimicrobials per kilogram of animal BW (combined weight of adult cows and PWC) per farm and ranged from 2.5 to 46.7 mg/kg of animal BW (Figure 1B). Herd-level mass density of antimicrobial summed for all herds was 545.2 mg/kg of BW. Ampicillin (30%) and ceftiofur (26%) accounted for more than half of combined antimicrobial mass density, followed by sulfadimethoxine (12%; Table3). Mass density of antimicrobials varied among active ingredients ($P < 0.001$; Table 3). When estimated using mass density, dihydrostreptomycin, ceftiofur, and ampicillin contributed the greatest mass, but did not differ from each other (Table 3; $P > 0.99$). Of farms that contained both PWC and adult cows, antimicrobials administered to adult cows represented 86% (95% CI: 80, 92) of the herd-level mass density of AMU and ranged from 36 to 99.9%.

For adult cows, mean mass density of antimicrobials was 12.1 ± 1.6 mg/kg of cow BW per farm and ranged from 1.6 to 36.3 mg/kg of cow BW. Mass density of antimicrobials used in cows in enrolled herds summed to 485.2 mg of antimicrobial per kilogram of adult cow BW. Among antimicrobials used in adult cows across routes, ampicillin and ceftiofur accounted for the greatest proportion of mass and represented 33% and 29% of antimicrobial mass given to adult cows, respectively.

For PWC, mean mass density of antimicrobials was 146 ± 40.6 mg/kg of PWC BW per farm and ranged from 1.0 to 1,075.1 mg/kg of PWC BW. Combined mass density for PWC in enrolled herds was 5,113.2 mg of antimicrobials per kilogram of PWC BW. Among antimicrobials given to PWC, greater proportion of mass was observed for sulfadimethoxine (28%) and trimethoprim-sulfa (29%; Table 6).

By Route of Administration in Adult Cows. When AMU was quantified using a mass-based metric, the greatest mass was contributed by antimicrobials given by an injectable route as compared with IMM (Table 5). The LSM mass density of antimicrobials given via an injectable route was 2 times greater than the mass density of IMM products (Table 5; $P < 0.001$). Proportionally, for adult cows, antimicrobials given by injection totaled 71% of antimicrobial mass density, while IMM (24%) and oral (6%) accounted for the remaining 29%.

Mean mass of antimicrobials used for IMM treatment of adult cows was 0.4 ± 0.05 mg/kg of cow BW per farm (Figure 2B) and ranged from 0.01 to 1.37 mg/kg. Combined mass of antimicrobials used for IMM treatment of lactating cows for enrolled herds summed to 16.1 mg/kg of adult cow BW. The LSM mass density of ceftiofur, cephapirin, and amoxicillin did not differ ($P = 0.99$), but greater proportion of mass was accounted for by ceftiofur (58% of all lactating cows IMM mg/kg of cow BW) and cephapirin (34% of all lactating cows IMM mg/kg of cow BW) (Table 4).

The mean mass density used for IMM treatment at dry off was 2.5 ± 0.3 mg/kg of cow BW per farm (Figure 2B) and ranged from 0.9 to 7.2 mg/kg of cow BW. The combined mass density of antimicrobials used for IMM treatment at dry off totaled 98.3 mg/kg of cow BW. No difference in LSM mass density was found among IMM antimicrobials given at dry off ($P = 0.37$), but the proportion of mass was mainly contributed by ceftiofur (36% of all IMM DCT mass) and dihydrostreptomycin (36% of all IMM DCT mass; Table 4).

Mean mass density for antimicrobials given systemically was 8.6 ± 1.4 mg of antimicrobials per kilogram of cow BW per farm (Figure 2B) and ranged from 0.2 to 32.8 mg/kg of cow BW. Combined mass density of antimicrobials administered systemically totaled 343.5 mg/kg of cow BW. Differences in mass were observed among antimicrobials used systemically (Table 4; $P < 0.001$), with ampicillin and ceftiofur representing the greatest mass. Among the 4 farms that used oral anti-

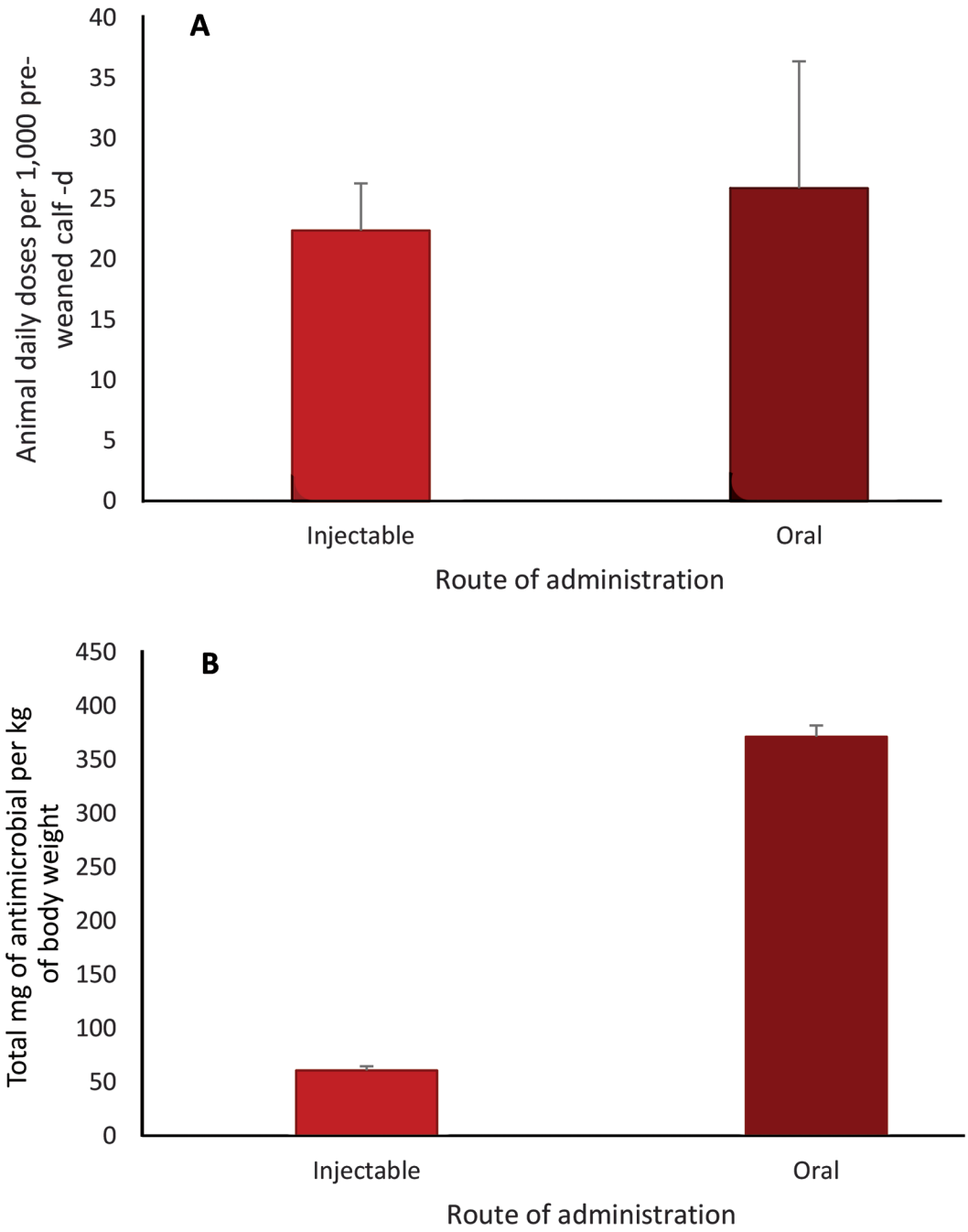

Figure 3. Antimicrobial usage in preweaned calves by administration route as estimated using dose-based (A) and mass-based (B) metrics. Bars represent SEM.

microbials, a total of 27.5 mg/kg of cow BW was quantified ($\bar{x} = 6.9 \pm 3.3$ mg/kg of cow BW).

By Route of Administration in PWC. When AMU was quantified for PWC using mass density, there was a large difference in LSM mass based on route of administration, but this difference was not statistically significant due to large variation in usage and a relatively small sample size (Table 5; $P = 0.30$).

Mean mass density of antimicrobials given by an injectable route was 61 ± 10.5 mg/kg of PWC BW (Figure 3B) and ranged from 1.0 to 272.8 mg/kg of PWC BW. The combined mass density of injectable antimicrobials totaled 2,139.4 mg/kg of PWC BW (Table 7). The mass of antimicrobials given to PWC varied among active ingredients (Table 7; $P < 0.001$). Ampicillin, enrofloxacin, florfenicol, gamithromycin,

Table 7. Animal daily doses (ADD) administered in preweaned calves (per 1,000 preweaned calf-d) via oral and injectable routes on 35 Wisconsin dairy farms

Active ingredient	Farm (n)	ADD per 1,000 preweaned calf-d					Mass-based (mg/kg of drug per BW)				
		LSM[1,2]	SEM	Median	Total	(%)[3]	LSM[1,2]	SEM	Median	Total	(%)[3]
Oral											
Oxytetracycline	1	—	—	9.99	9.99	4.82	—	—	160.47	160.47	5.40
Sulfadimethoxine	2	—	—	48.53	97.06	46.86	—	—	676.19	1,352.39	45.48
TMP-sulfa[4]	5	—	—	24.01	100.06	48.31	—	—	350.62	1,460.88	49.13
Injectable											
Ampicillin	16	1.32[ab]	1.61	2.46	67.11	8.56	5.20[a]	1.60	9.89	269.46	12.60
Ceftiofur	20	0.57[ab]	1.54	0.52	90.95	11.60	0.66[b]	1.53	0.53	83.21	3.89
Danofloxacin	1	—	—	0.86	0.86	0.11	—	—	1.89	1.89	0.09
Enrofloxacin	27	1.34[ab]	1.45	1.74	106.24	13.56	3.94[a]	1.45	4.70	270.12	12.63
Florfenicol	21	0.51[ab]	1.52	0.53	42.96	5.48	6.75[a]	1.51	6.96	576.17	26.93
Gamithromycin	11	0.44[b]	1.77	0.71	14.97	1.91	1.05[ab]	1.76	1.68	35.27	1.65
Gentamicin	2	—	—	0.09	0.17	0.02	—	—	0.38	0.77	0.04
Oxytetracycline	10	0.48[ab]	1.82	1.48	34.08	4.35	2.85[ab]	1.80	8.75	205.20	9.59
Penicillin G	24	3.76[a]	1.48	3.09	315.32	40.23	5.54[a]	1.48	4.52	460.36	21.52
Sulfadimethoxine	1	—	—	5.81	5.81	0.74	—	—	69.96	69.96	3.27
Tildipirosin	4	—	—	0.2	0.99	0.13	—	—	0.29	1.45	0.07
Tilmicosin	5	—	—	0.41	2.16	0.28	—	—	2.98	15.79	0.74
Tulathromycin	30	0.80[ab]	1.43	0.7	102.05	13.02	0.80[b]	1.42	0.67	149.44	6.99
Tylosin	1	—	—	0.05	0.05	0.01	—	—	0.33	0.33	0.02

[a,b]Mean values within the same column with different superscripts differ from each other ($P < 0.05$).

[1]Active ingredients containing 5 or less farms were not used in the comparison among means.

[2]Statistical analyses were performed on natural logs for farms using the active ingredient; data are presented are back transformed LSM.

[3]Proportion within route of antimicrobial usage.

[4]Trimethoprim (TMP)-sulfamethoxazole combination.

and oxytetracycline had the greatest mass density and did not differ among each other ($P > 0.99$).

Mean mass density for antimicrobials given orally was 371.7 ± 126.4 mg/kg of PWC BW per farm (Figure 3B) and ranged from 3.9 to 1,060.3 mg/kg of PWC BW. The combined mass density of oral antimicrobials used for these herds totaled 2,973.7 mg/kg of PWC BW (Table 7). Three antimicrobials (oxytetracycline, sulfadimethoxine, and trimethoprim-sulfa) were used for oral administration, with 95% of the usage attributed to sulfadimethoxine (46%) and trimethoprim-sulfa (49%).

DISCUSSION

Antimicrobial usage on US dairy farms has previously been quantified on smaller farms in Wisconsin (Pol and Ruegg, 2007) and Pennsylvania (Redding et al., 2019), as well as in countries such as Canada (Saini et al., 2012), Belgium (Stevens et al., 2016), Austria (Firth et al., 2017), the United Kingdom (Hyde et al., 2017), and Argentina (González Pereyra et al., 2015). All previous studies have enrolled small or midsized farms, and none have focused on large farms. Although we included only 40 herds, our farms contained >52,000 cows, thus providing a large number of animals who would be potentially susceptible to bacterial diseases and may benefit from antimicrobial therapy. Inclusion of large farms adds valuable perspective about AMU on dairy farms that produce the majority of milk in the United States (MacDonald and Newton, 2014). In addition, our use of a dose-based metric allowed comparison to previous studies, and quantification using a mass-based metric provided important new information that illustrated how choice of metric can influence interpretation of AMU.

Enrolled dairy farms represented about 10% of all Wisconsin dairy farms with >250 dairy cows and were representative of this demographic as they used typical management practices for larger herds in this region (USDA–APHIS–VS–CEAH–NAHMS, 2014; Rowbotham and Ruegg, 2015). Farms were recruited based on herd size and availability of treatment records, and they likely represented dairy herds that have better recording systems than the overall population of Wisconsin dairy herds (Hoe and Ruegg, 2006). This trend was previously observed by USDA–APHIS–VS–CEAH (2007) researchers, who reported that adoption of computerized dairy management records increased from about 9% of small farms (<100 cows) to 38% of the medium farms (100–499 cows) and 83% of large farms (>500 cows). Although selection criteria included questions about availability of records, some data were missing for some herds, including name of drug, number of days treated, or dosage administered, as has been noted in previous publications (Wenz and Giebel, 2012). The

most popular dairy management software provides only 8 characters to describe an event, which limits opportunities for full recording of some treatments. In the United States, dairy farm records are private, and in contrast to some other countries (Espetvedt et al., 2013), there is no centralized registry for treatment records of agricultural animals. In our study, all farms recorded AMU data in computerized recording systems and farmers were interviewed to verify disease definitions and treatment protocols. The 1-yr retrospective data collection period allowed us to capture seasonal effects that may influence AMU (Mills et al., 2018). Review of computerized health records was useful as we were able to assess the number of treatments per disease as well as the number of days each treatment was administered. A minority of disease events were not entered in electronic records, and these events and the proportion of nonrecorded cases receiving antimicrobials were estimated by farm owners during the survey. The possibility of information bias cannot be excluded because, in some instances, we interviewed farm owners who did not personally administer antimicrobials, and there may have been errors in administering treatment protocols. Even though >90% of data were obtained from electronic dairy management records, it is possible that recall or recording bias could have influenced our results.

Ceftiofur and cephapirin were the most common antimicrobials used on these farms, and ceftiofur was the only antimicrobial used on all farms. Several products and routes are used to administer ceftiofur, and this antimicrobial accounted for almost half of herd-level ADD, more than half of cow-level ADD, and 9% of ADD used in PWC. Ceftiofur has previously been reported as the most frequent antimicrobial used for treatment of diseases other than mastitis, with 85% of smaller farms using ceftiofur to treat respiratory problems and metritis, and 65% of smaller farms using ceftiofur to treat foot infections (Pol and Ruegg, 2007). In Canada, ceftiofur represented 15% of total ADD used at the national level, and usage increased with increasing herd size (Saini et al., 2012). Use of ceftiofur as an IMM antimicrobial is approved in the United States, but some nonapproved IMM use of ceftiofur to treat mastitis was reported even before approval of the IMM ceftiofur product (FDA, 2005; Pol and Ruegg, 2007). In the United States, several formulations of ceftiofur are approved for use in dairy cattle, and none of the systemically administered products require a milk withholding period when used according to label indications, dosage, and route. Administration of ceftiofur outside of approved label instructions for dose, frequency, duration, or route is not permitted by FDA regulations, but extralabel usage of the label dose, frequency, and duration is allowed for treatment of conditions for which no current antimicrobials have an approved label indication. The approved IMM ceftiofur product is very popular and has been used to treat about half of all cases of clinical mastitis in the United States (USDA–APHIS–VS–CEAH, 2008a; USDA–APHIS–VS–CEAH–NAHMS, 2014). The popularity of this product is probably based on several favorable characteristics, including broad-spectrum activity, a dosing schedule that includes once daily administration, and a flexible dosing interval of 2 to 8 d.

Of 7 IMM antimicrobials approved for administration to dry cows, 5 are β-lactams, and the other 2 products include the only antimicrobial combination products licensed for use in US dairy cows. Only 2 classes of antimicrobials (6 β-lactam products and 1 lincosamide) are approved for IMM treatment of lactating cows in the United States, and all but 1 of those products were used by farms enrolled in our study. Similar to previous studies (Pol and Ruegg, 2007; Redding et al., 2019), first and third generation cephalosporins were used most frequently and accounted for the greatest proportion of ADD and of mass for IMM treatment of lactating cows. Use of ceftiofur is concerning because the World Health Organization has classified third generation cephalosporins as a critically important class for human health (WHO, 2018a), and responsible usage guidelines encourage use of narrow spectrum antimicrobials when appropriate.

Among the 18 active ingredients reported for injectable usage in adult cows, 2 antimicrobials are not approved for usage in adult cows and accounted for a small proportion of total AMU in adult cows. For PWC, the main classes used were macrolides (gamithromycin, tildipirosin, tilmicosin, tulathromycin, and tylosin), fluoroquilones (danofloxacin and enroxoflacin), penicillins (ampicillin and penicillin G), amphenicols (florfenicol), cephalosporins (ceftiofur), tetracyclines (oxytetracycline), and sulfonamides. Our estimates are comparable with USDA survey data that indicated that macrolides and amphenicols were the primary antimicrobials used for treatment of respiratory diseases in PWC and reinforced that tetracyclines, ceftiofur, and trimethoprim-sulfa are the primary antimicrobials used for treatment of digestive diseases (USDA–APHIS–VS–CEAH–NAHMS, 2018).

Several metrics have been used for measurement of AMU, and the choice of metric should be based on the purpose for measuring AMU (Mills et al., 2018). Our data demonstrated that interpretation of AMU can be altered depending on the metric that is used. None of the metrics are ideal for all situations, but measurement of AMU is essential to evaluate interventions used to reduce AMU and for research about

potential associations of AMU with measures of AMR. Regardless of metric, accuracy and consistency of the health records are crucial for analysis of AMU (Wenz and Giebel, 2012). When quantifying AMU, animal weight is an important component (Mills et al., 2018) as it may vary among farms and breeds, thus altering calculations. For example, in our study, 4 breeds were reported, but Holsteins represented 99% of total cows. During the farm visit, we asked a question regarding animal weight. Owner- and manager-estimated weights were very similar among farms, and thus we used a standard BW for calculating the mass-based metric for all farms.

We used ADD as our dose-based metric because this standardized method is widely adopted for quantification of AMU in both humans and animals (Jensen et al., 2004; CDC, 2014; WHO, 2018b). In some countries, ADD has been used to measure changes in AMU as the result of national legislative efforts, but comparisons among countries is complicated by differences in how the metric is calculated (Taverne et al., 2015). The formula used to calculate ADD can be altered to demonstrate larger or smaller reductions in AMU. For example, in dairy herds, some studies have considered 4 IMM dry cow antimicrobial tubes as 4 doses (Stevens et al., 2016), whereas others have considered the 4 doses to be a single ADD (Pol and Ruegg, 2007; Redding et al., 2019). When the goal is to reduce ADD on dairy farms, selective DCT programs are often pursued (Scherpenzeel et al., 2016) and use of a single (rather than 4) IMM tube to define the standard ADD will have the effect of magnifying apparent reductions in AMU. The effect of using 4 versus 1 IMM tubes as an ADD is apparent when comparing results from Pol and Ruegg, (2007), where IMM treatment of clinical mastitis accounted for more AMU than DCT versus our current study, where we observed that DCT accounted for the majority of IMM AMU. Most previous studies that measured AMU on dairy farms have used ADD, although denominators have varied (Pol and Ruegg, 2007; Saini et al., 2012; Stevens et al., 2016; Redding et al., 2019). Use of ADD allows comparisons among different antimicrobials without regard for potency, concentration, units, or route (WHO, 2018b). One disadvantage of ADD is that it does not account for multiple administrations per day, and ADD may vary depending on the approved dosing schedule among countries. When using ADD to quantify antimicrobial usage, variation in BW among herds should be clearly specified, as well as the minimum, mean, or maximum dose rate chosen to be the defined daily dose because these choices significantly affect final assessments of AMU (Mills et al., 2018).

Although farms enrolled in our study were in the same state and shared many management characteristics, considerable variation in AMU was observed among herds. Use of different metrics influenced overall ranking of AMU, but higher-consuming herds remained in the upper quartiles, regardless of metric. Compared with the herd that used the least ADD, the herd that was ranked highest used about 7 times more ADD per 1,000 animal-d. Both of the herds that recorded the greatest number of ADD used considerable antimicrobials to treat PWC. This study was not designed to investigate management practice risk factors associated with AMU and AMR, but wide variation in AMU among herds demonstrated that there is considerable opportunity for reductions in AMU based on adoption of management practices that are already used by herds consuming fewer doses.

Herd-level comparisons of ADD among studies are difficult because of differences in approved product labels among countries and variation in denominators. Most previous research has quantified AMU only for adult cows (Pol and Ruegg, 2007; Stevens et al., 2016; Hyde et al., 2017), but few have included calves (González Pereyra et al., 2015; Redding et al., 2019). When ADD is calculated using only PWC in the denominator, the smaller number of calf days results in about 1.5 times greater ADD per calf day as compared with ADD density measured in cows. This is expected because adult dairy cows are typically at risk for diseases that are treated with antimicrobials for a relatively short proportion of the typical 365-d lactation cycle, and the need to discard milk results is an economic disincentive for treatment of lactating cows. In contrast, PWC are vulnerable to infectious bacterial diseases for most of the period before weaning, and they are expected to remain in the herd for years, thus meat and withholding periods are not as great of a concern.

Some studies have quantified AMU using ADD per cow per year (Pol and Ruegg, 2007; González Pereyra et al., 2015), and ADD density (ADD per 1,000 cow-d) can be converted to this value by dividing by 2.74 (1,000 cow-d/365 d). When our data are converted [(15.8 ADD/1,000 cow-d)/2.74 = 5.8 ADD/cow per year], AMU as measured by ADD is remarkedly similar to previous estimates for smaller herds in Wisconsin (5.4 ADD/cow per year; (Pol and Ruegg, 2007) and Canada (5.2 ADD per cow per year; (Saini et al., 2012). As reported in previous studies (Pol and Ruegg, 2007; González Pereyra et al., 2015; Hyde et al., 2017; Redding et al., 2019), when measured using ADD, IMM administration has a considerable effect on estimates of AMU. In our study, use of IMM antimicrobials accounted for 78% of ADD given to adult cows. Mastitis

is the most frequent bacterial disease occurring on dairy farms, and until recently, antimicrobials were generally administered based on clinical signs, regardless of etiology (Ruegg, 2017), and blanket DCT has been routinely recommended as part of a mastitis control program (NMC, 2020). As selective dry cow and lactating cow therapy programs for clinical mastitis are increasingly adopted (Ruegg, 2018), it is likely that AMU attributable to IMM therapy on dairy farms will decline.

As compared with ADD, the mass-based estimate was easier to calculate, but this metric does not account for variation in potency among active ingredients, and thus favors antimicrobials with lower dosing concentrations (e.g., ceftiofur). One previous study compared mass-based and dose-based metrics (Hyde et al., 2017; Redding et al., 2019), and similar to our study, they demonstrated that the predominant route and antimicrobials responsible for the greatest proportion of usage are altered by use of different metrics. Depending on metric, dramatic differences in usage are inferred based on route. When ADD is used, IMM administration accounts for about 78% of doses, but when mass density is used, the proportions are reversed and systemic administration totals 71% of mass. The significance of selecting a metric relative to studying associations between AMU and development of AMR are unknown and should be explored in future studies.

As compared with estimating AMU in adult cows, quantification of AMU in replacement animals is more difficult. Issues for estimating AMU in replacements include larger variation in BW among youngstock (González Pereyra et al., 2015), transfer of replacement animals to remote facilities (rather than remaining on site), and maintenance of fewer treatment records for this group of animals (Zwald et al., 2004). It is apparent from our data that some farms used considerable quantities of antimicrobials for treatment of PWC, and use of oral antimicrobials accounted for 21% of ADD and 58% of mass. Products given via oral administration are often given in greater dosages, thus increasing the total exposure to active ingredients. Reduced resistance of commensal *Escherichia coli* to some antimicrobials has been documented with reduced antimicrobial usage in calves (Afema et al., 2019), and increased education of farmers relative to AMU in this class of animals is needed.

CONCLUSIONS

Antimicrobial usage in large Wisconsin dairy farms was estimated using 2 different metrics. When estimated using ADD, AMU in adult cows was similar to previous reports that included smaller farms and different countries. Regardless of metric, AMU varied substantially among farms, and future studies should identify risk factors associated with greater consumption of antimicrobials. Ceftiofur was used in multiple formulations and accounted for a large proportion of AMU. At the herd level, adult cows represented the greatest proportion of AMU due to the greater number of animals in this category, but greater mass density of AMU was observed for PWC. At the cow level, IMM administration was the primary route of AMU when ADD was used for estimation, but systemic administration accounted for most antimicrobial usage when mass density was used for estimation. Among antimicrobials used on adult cows, cephalosporins were most frequently administered and represented a significant proportion of AMU, regardless of metric. Among antimicrobials administered to PWC, β-lactams and macrolides were the primary classes of antimicrobials. Overall, these results show that choice of metric and consideration of route of administration can substantially influence estimates of AMU on large dairy farms.

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REFERENCES

Afema, J. A., M. A. Davis, and W. M. Sischo. 2019. Antimicrobial use policy change in pre-weaned dairy calves and its impact on antimicrobial resistance in commensal *Escherichia coli*: A cross sectional and ecological study. BMC Microbiol. 19:217. https://doi.org/10.1186/s12866-019-1576-6.

Aiello, S. E., M. A. Moses, and D. G. Allen. 2016. Pharmacology. Pages 2666–2692 in The Merck Veterinary Manual. Eleventh edition. Merck & Co., Inc, Kenilworth, NJ.

Aminov, R. I. 2010. A brief history of the antibiotic era: Lessons learned and challenges for the future. Front. Microbiol. 1:134. https://doi.org/10.3389/fmicb.2010.00134.

CDC. 2014. Core Elements of Hospital Antibiotic Stewardship Programs. Accessed Jun. 7, 2020. https://www.cdc.gov/antibiotic-use/core-elements/hospital.html.

CDC. 2017. Antibiotic Use in the United States, 2017: Progress and Opportunities. Accessed Jun. 7, 2020. https://www.cdc.gov/antibiotic-use/stewardship-report/pdf/stewardship-report.pdf.

Davies, J., and D. Davies. 2010. Origins and evolution of antibiotic resistance. Microbiol. Mol. Biol. Rev. 74:417–433. https://doi.org/10.1128/MMBR.00016-10.

Espetvedt, M. N., O. Reksen, S. Rintakoski, and O. Østerås. 2013. Data quality in the Norwegian dairy herd recording system: Agreement between the national database and disease recording on farm. J. Dairy Sci. 96:2271–2282. https://doi.org/10.3168/jds.2012-6143.

FARAD. 2021. Special Topics: The Use of Tulathromycin and Florfenicol in Dairy. Accessed Jan. 27, 2021. http://www.usfarad.org/special-topics.html.

FDA. 2005. Federal Register- Rules and Regulations. Accessed Oct. 8, 2019. https://www.regulations.gov/document?D=FAA-2005-20424-0001.

Firth, C. L., A. Käsbohrer, C. Schleicher, K. Fuchs, C. Egger-Danner, M. Mayerhofer, H. Schobesberger, J. Köfer, and W. Obritzhauser.

2017. Antimicrobial consumption on Austrian dairy farms: An observational study of udder disease treatments based on veterinary medication records. PeerJ 5:e4072. https://doi.org/10.7717/peerj.4072.

Grave, K., C. Greko, L. Nilsson, K. Odensvik, T. Mørk, and M. Rønning. 1999. The usage of veterinary antibacterial drugs for mastitis in cattle in Norway and Sweden during 1990–1997. Prev. Vet. Med. 42:45–55. https://doi.org/10.1016/S0167-5877(99)00057-4.

Hao, H., G. Cheng, Z. Iqbal, X. Ai, H. I. Hussain, L. Huang, M. Dai, Y. Wang, Z. Liu, and Z. Yuan. 2014. Benefits and risks of antimicrobial use in food-producing animals. Front. Microbiol. 5:288. https://doi.org/10.3389/fmicb.2014.00288.

Hoe, F. G. H., and P. L. Ruegg. 2006. Opinions and practices of Wisconsin dairy producers about biosecurity and animal well-being. J. Dairy Sci. 89:2297–2308. https://doi.org/10.3168/jds.S0022-0302(06)72301-3.

Hyde, R. M., J. G. Remnant, A. J. Bradley, J. E. Breen, C. D. Hudson, P. L. Davies, T. Clarke, Y. Critchell, M. Hylands, E. Linton, E. Wood, and M. J. Green. 2017. Quantitative analysis of antimicrobial use on British dairy farms. Vet. Rec. 181:683. https://doi.org/10.1136/vr.104614.

Jensen, V. F., E. Jacobsen, and F. Bager. 2004. Veterinary antimicrobial-usage statistics based on standardized measures of dosage. Prev. Vet. Med. 64:201–215. https://doi.org/10.1016/j.prevetmed.2004.04.001.

Johnston, A. M. 1998. Use of antimicrobial drugs in veterinary practice. BMJ 317:665–667. https://doi.org/10.1136/bmj.317.7159.665.

Jones, C. M., and J. Heinrichs. 2020. Growth Charts for Dairy Heifers. Accessed Jan. 27, 2021. https://extension.psu.edu/growth-charts-for-dairy-heifers#section-2.

MacDonald, J., and D. Newton. 2014. Milk Production Continues Shifting to Large-Scale Farms. Accessed Jun. 7, 2020. https://www.ers.usda.gov/amber-waves/2014/december/milk-production-continues-shifting-to-large-scale-farms/.

MacFadden, D. R., D. Fisman, J. Andre, Y. Ara, M. S. Majumder, I. I. Bogoch, N. Daneman, A. Wang, M. Vavitsas, L. Castellani, and J. S. Brownstein. 2016. A platform for monitoring regional antimicrobial resistance, using online data sources: Resistanceopen. J. Infect. Dis. 214(Suppl. 4):S393–S398. https://doi.org/10.1093/infdis/jiw343.

McGuirk, S. Sick Calf Protocols. Accessed Mar. 5, 2018. https://www.vetmed.wisc.edu/dms/fapm/fapmtools/8calf/calf_protocols_ver4.pdf.

Mills, H. L., A. Turner, L. Morgans, J. Massey, H. Schubert, G. Rees, D. Barrett, A. Dowsey, and K. K. Reyher. 2018. Evaluation of metrics for benchmarking antimicrobial use in the UK dairy industry. Vet. Rec. 182:379. https://doi.org/10.1136/vr.104701.

NMC. 2020. Recommended Mastitis Control Program. Accessed Jun. 7, 2020. https://www.nmconline.org/documents/.

González Pereyra, V., M. Pol, F. Pastorino, and A. Herrero. 2015. Quantification of antimicrobial usage in dairy cows and preweaned calves in Argentina. Prev. Vet. Med. 122:273–279. https://doi.org/10.1016/j.prevetmed.2015.10.019.

Pol, M., and P. L. Ruegg. 2007. Treatment practices and quantification of antimicrobial drug usage in conventional and organic dairy farms in Wisconsin. J. Dairy Sci. 90:249–261. https://doi.org/10.3168/jds.S0022-0302(07)72626-7.

Redding, L. E., J. Bender, and L. Baker. 2019. Quantification of antibiotic use on dairy farms in Pennsylvania. J. Dairy Sci. 102:1494–1507. https://doi.org/10.3168/jds.2018-15224.

Rowbotham, R. F., and P. L. Ruegg. 2015. Association of bedding types with management practices and indicators of milk quality on larger Wisconsin dairy farms. J. Dairy Sci. 98:7865–7885. https://doi.org/10.3168/jds.2015-9866.

Ruegg, P. L. 2017. A 100-year review: Mastitis detection, management, and prevention. J. Dairy Sci. 100:10381–10397. https://doi.org/10.3168/jds.2017-13023.

Ruegg, P. L. 2018. Making antibiotic treatment decisions for clinical mastitis. Vet. Clin. North Am. Food Anim. Pract. 34:413–425. https://doi.org/10.1016/j.cvfa.2018.06.002.

Saini, V., J. T. McClure, D. Léger, S. Dufour, A. G. Sheldon, D. T. Scholl, and H. W. Barkema. 2012. Antimicrobial use on Canadian dairy farms. J. Dairy Sci. 95:1209–1221. https://doi.org/10.3168/jds.2011-4527.

Scherpenzeel, C. G. M., I. E. M. Den Uijl, G. Van Schaik, R. G. M. Olde Riekerink, J. M. Keurentjes, and T. J. G. M. Lam. 2014. Evaluation of the use of dry cow antibiotics in low somatic cell count cows. J. Dairy Sci. 97:3606–3614. https://doi.org/10.3168/jds.2013-7655.

Scherpenzeel, C. G. M., I. E. M. den Uijl, G. van Schaik, R. G. M. O. Riekerink, H. Hogeveen, and T. J. G. M. Lam. 2016. Effect of different scenarios for selective dry-cow therapy on udder health, antimicrobial usage, and economics. J. Dairy Sci. 99:3753–3764. https://doi.org/10.3168/jds.2015-9963.

Stevens, M., S. Piepers, K. Supré, J. Dewulf, and S. De Vliegher. 2016. Quantification of antimicrobial consumption in adult cattle on dairy herds in Flanders, Belgium, and associations with udder health, milk quality, and production performance. J. Dairy Sci. 99:2118–2130. https://doi.org/10.3168/jds.2015-10199.

Taverne, F. J., J. H. Jacobs, D. J. J. Heederik, J. W. Mouton, J. A. Wagenaar, and I. M. van Geijlswijk. 2015. Influence of applying different units of measurement on reporting antimicrobial consumption data for pig farms. BMC Vet. Res. 11:250. https://doi.org/10.1186/s12917-015-0566-7.

USDA–APHIS–VS–CEAH. 2007. Dairy 2007, Part I: Reference of Dairy Cattle Health and Management Practices in the United States, 2007. Accessed Jul. 6, 2020. https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/monitoring-and-surveillance/nahms/nahms_dairy_studies.

USDA–APHIS–VS–CEAH. 2008a. Antibiotic Use on U.S. Dairy Operations, 2002 and 2007. Accessed Jun. 7, 2020. https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/monitoring-and-surveillance/nahms/nahms_dairy_studies.

USDA–APHIS–VS–CEAH. 2008b. Dairy 2007, Part III: Reference of Dairy Cattle Health and Management Practices in the United States. Accessed Jul. 6, 2020. https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/monitoring-and-surveillance/nahms/nahms_dairy_studies.

USDA–APHIS–VS–CEAH–NAHMS. 2014. Dairy 2014, Milk Quality, Milking Procedures, and Mastitis on US Dairies, 2014. Accessed Jun. 7, 2020. https://www.aphis.usda.gov/animal_health/nahms/dairy/downloads/dairy14/Dairy14_dr_Mastitis.pdf.

USDA–APHIS–VS–CEAH–NAHMS. 2018. Dairy 2014 Health and Management Practices on U.S. Dairy Operations, 2014. Accessed Jun. 7, 2020. https://www.aphis.usda.gov/animal_health/nahms/dairy/downloads/dairy14/Dairy14_dr_PartIII.pdf.

Wade, O. 1984. Drug utilization studies—The first attempts. Plenary lecture. In: Drug Utilization Studies: Implications for Medical Care. F. Sjöqvist and I. Agenäs, ed. Acta Medica Scandinavica. (Suppl.) 683:7–9.

Wenz, J. R. R., and S. K. K. Giebel. 2012. Retrospective evaluation of health event data recording on 50 dairies using Dairy Comp 305. J. Dairy Sci. 95:4699–4706. https://doi.org/10.3168/jds.2011-5312.

WHO. 2003. Introduction to Drug Utilization Research. Accessed Jun. 7, 2020. https://apps.who.int/iris/handle/10665/42627.

WHO. 2018a. Critically Important Antimicrobials for Human Medicine: 6th Revision. Accessed Jul. 6, 2020. https://apps.who.int/iris/bitstream/handle/10665/312266/9789241515528-eng.pdf.

WHO. 2018b. WHO Collaborating Centre for Drug Statistics Methodology, Guidelines for ATC Classification and DDD Assignment 2019. Accessed Jun. 7, 2020. https://www.whocc.no/atc_ddd_index_and_guidelines/guidelines/.

Zwald, A. G., P. L. Ruegg, J. B. Kaneene, L. D. Warnick, S. J. Wells, C. Fossler, and L. W. Halbert. 2004. Management Practices and Reported Antimicrobial Usage on Conventional and Organic Dairy Farms. J. Dairy Sci. 87:191–201. https://doi.org/10.3168/jds.S0022-0302(04)73158-6.