Observational Cohort Study of the Safety of Digoxin Use in Women with Heart Failure

Journal:	*BMJ Open*
Manuscript ID:	bmjopen-2012-000888
Article Type:	Research
Date Submitted by the Author:	16-Jan-2012
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Primary Subject Heading:	Cardiovascular medicine
Secondary Subject Heading:	Epidemiology, Evidence based practice
Keywords:	Cardiac Epidemiology < CARDIOLOGY, Heart failure < CARDIOLOGY, EPIDEMIOLOGY

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Keywords: heart failure, digoxin, mortality, sex

Support: *FOCUS Medical Student Fellowship in Women's Health* supported by the Edna G. Kynett Memorial Foundation, Pharmacoepidemiology Training Funds donated to the Center for Clinical Epidemiology and Biostatistics of the University of Pennsylvania School of Medicine. The study sponsors had no influence over design of the study, the handling of data, the composition of the manuscript, or the decision to submit it.

All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author). SH has consulted for Abbott Laboratories. BLS has received grant funding from Abbott Laboratories, payment for lectures from Teva Pharmaceutical Industries, has consulted for Teva and for GlaxoSmithKline. All authors on this manuscript have a connection to a training program which has received funding from Abbott Laboratories and GlaxoSmithKline. None of these connections related directly to digoxin. These companies are among the over 50 corporations that manufacture or distribute digoxin.

Guarantorship and Contributorship: JHF is the guarantor of this manuscript, having been primarily responsible for study design, data analysis including programming and biostatistics, and manuscript composition. JHF, BK, KH, SB, JM, CR, BLS , and SH all made substantial contributions to the conception and design of this study. JHF was primarily responsible for analysis and interpretation of the data but all listed authors made significant contributions to this as well. All of the listed authors made substantial contributions in drafting the article or revising it critically for important intellectual content.

SUMMARY

Objectives This study aims to assess whether digoxin has a different effect on mortality risk for women than it does for men in patients with heart failure (HF).

Design This study uses the United Kingdom-based The Health Information Network (THIN) population database in a cohort study of the impact of digoxin exposure on mortality for men and women who carry the diagnosis of HF. Digoxin exposure was assessed based on prescribing data. Multivariable Cox proportional hazards models were used to assess whether there was an interaction between sex and digoxin affecting mortality hazard.

Setting The setting was primary care outpatient practices.

Participants The study cohort consisted of 17,707 men and 19,227 women with the diagnosis of HF who contributed only time without digoxin exposure and 9,487 men and 10,808 women with the diagnosis of HF who contributed time with digoxin exposure.

Main Outcome Measures The main outcome measure was all-cause mortality.

Results The primary outcome of this study was the absence of a large interaction between digoxin use and sex affecting mortality. For men digoxin use was associated with a hazard ratio for mortality of 1.00, while for women the hazard ratio was also 1.00 (p-value for interaction 0.65). Sensitivity analyses did not affect this estimate materially.

Conclusion Observational data do not support the concern that there is a substantial increased risk of mortality due to the use of digoxin in women. This finding is consistent with previous observational studies but discordant with results from a post-hoc analysis of a randomized controlled trial of digoxin versus placebo.

INTRODUCTION

It has been hypothesized that digoxin, when used in the treatment of heart failure (HF), may increase mortality by approximately 20% in women but not in men. This hypothesis is based on a post-hoc analysis in 6800 patients by Rathore and colleagues of the Digitalis Investigation Group (DIG) trial, a placebo-controlled randomized study that showed digoxin did not affect overall mortality but did reduce hospitalizations in patients with HF.[1] The post-hoc analysis examined mortality effects by sex and found that compared to placebo digoxin conferred reduced mortality in men (absolute difference -1.6%, 95% CI -4.2% to 1.0%) and increased mortality in women (absolute difference 4.2%, 95% CI -0.5% to 8.8%), with a statistically significant interaction (p value= 0.034).[2] One proposed mechanism is that women may have higher mean serum digoxin levels than men; there is evidence that even within the therapeutic range higher serum digoxin levels are associated with increased mortality.[3,4]

Because the sex-digoxin interaction was based on post-hoc analyses, it needs cautious interpretation. Yet, if true, the finding is clinically important because digoxin continues to be widely used by both women and men.[5] It remains unclear whether digoxin should be used differently in the different sexes, and concerns about its use in women continue to appear in the literature.[6,7] Unwarranted recommendations against use of digoxin in women would deprive a large population of a medicine with demonstrated ability to prevent hospitalizations and, by implication, improve quality of life.

Further randomized trials evaluating the interaction between digoxin and sex have not emerged. One observational study based on the Studies of Left Ventricular Dysfunction (SOLVD) cohort found no difference in digoxin's effect on mortality between sexes, but concluded that additional research in other populations is still needed.[7] Limitations of that study included restriction to a population with severely reduced ejection fraction (<35%) and relatively small sample size (n=6797). We sought to conduct a much larger study in a broader population that could help assess whether digoxin increases mortality in women compared to men.

METHODS

Study Design: We conducted a retrospective cohort study to test the primary hypothesis that sex was an effect modifier for the association of digoxin use with mortality. We also conducted planned secondary analyses of whether effect modification was mediated by digoxin dose or by serum digoxin concentration. This study was approved by the Institutional Review Board of the University of Pennsylvania. Analyses were conducted using SAS 9.1.

Study Population: The study used the Health Improvement Network (THIN) database, a primary care electronic medical record database in the UK. This database contains over 5 million individuals who have contributed person-time from over 300 different general practices from 1986 to 2008. THIN includes demographic information on patients, as well as records of prescribed drugs, medical diagnoses (coded as READ codes), as well as vital signs and laboratory values on a subset of patients. It is a representative subset of the United Kingdom's general population.[8]

Cohort Definition: The analysis was restricted to individuals with a diagnostic code for HF. Once that condition was met, patients could contribute follow-up time as long as they were being given regular prescriptions of at least one drug consistent with the treatment for HF (as described below).

Information was only used if it was collected during times when the adjusted mortality ratio, a quality control measure used in THIN to identify practices which are recording deaths accurately, was within acceptable standards for a given practice.[9] It was possible for one individual to contribute both exposed and unexposed time. The result was four cohorts with some within-sex crossover: men on digoxin, men not on digoxin, women on digoxin, women not on digoxin.

Exposed Group: Exposed subjects were defined as individuals who carried the diagnosis of HF and were receiving multiple consecutive prescriptions for digoxin. Individuals were assumed to be on digoxin from the date of receipt of a prescription until 30 days after their prescription ran out (the intended duration of a prescription was either computed from information on daily dose and number of tablets, or imputed as 30 days if that information was not available).

Unexposed Group: Unexposed subjects were defined as individuals who carried a diagnosis of HF and were receiving regular prescriptions for a loop diuretic, beta blocker, angiotensin converting enzyme (ACE) inhibitor, or angiotensin receptor blocker (ARB). These are drugs that, while not specific for HF, are commonly used for that condition.

Definition of Outcome: The outcome for this study was all-cause mortality, as documented by a date of death in the THIN demographics file.

Definition of Covariates: Baseline covariates consisted of diagnostic codes for common comorbidities of HF, including hypertension, stroke, myocardial infarction (MI), chronic obstructive pulmonary disease (COPD), diabetes, and atrial fibrillation. We also assessed age, sex, and baseline use of drugs commonly used in HF, notably aspirin, statins, oral anticoagulants, potassium-sparing diuretics, and the HF-associated drugs listed above. To be included as a baseline variable drug use had to be in the year prior to HF diagnosis; medical history such as a history of hypertension could be coded at any time prior to HF diagnosis. Blood pressure, body mass index (BMI), and a variety of lab values including serum creatinine were available on a minority of participants and were used as baseline variables in sensitivity analysis. Once a patient began to contribute exposure-time, baseline variables were no longer updated.

Statistical Analysis: Because of the large sample size, we anticipated that even clinically insignificant differences between baseline variables could be statistically significant and therefore described baseline variables using standardized differences.[10] Standardized differences are a balance diagnostic used to assess how similar groups are at baseline – conceptually they are equal to the difference of a variable's mean between two groups divided by the standard deviation for that variable. A standardized difference of < 0.1, by convention, is considered to indicate good balance of groups on that variable.[10] For categorical variables a standardized difference between men and women was calculated as $\frac{p_{women}-p_{men}}{\sqrt{\left(\frac{p_{women}(1-p_{women})+p_{men}(1-p_{men})}{2}\right)}}$, where p = proportion of the population falling into a category. In addition, because the most relevant question was whether these between-sex differences were consistent in both digoxin-exposed and digoxin-unexposed groups, this equation was extended to treat the inter-sex differences for the exposed and unexposed groups as variables which in turn had their own standardized differences:

$$[(p_{women,exposed} - p_{men,exposed}) - (p_{women,unexposed} - p_{men,unexposed})]/\sqrt{[(p_{women,exposed}(1-p_{women,exposed}) + p_{men,exposed}(1-p_{men,exposed}) + p_{women,unexposed}(1-p_{women,unexposed}) + p_{men,unexposed}(1-p_{men,unexposed}))/4]}$$

Total follow-up time for all individuals was assessed, with digoxin use, digoxin dose, and serum digoxin concentrations as time-varying covariates in secondary analysis. In the primary analysis digoxin dose and serum concentration were ignored. A Cox proportional hazards model was used to calculate hazard ratios for sex, digoxin use, and the interaction of sex and digoxin use, controlling for the covariates specified above.[11]

The primary parameter of interest was the adjusted hazard ratio for the interaction of sex and digoxin use, where a significant deviation from the null would indicate that digoxin use was associated with mortality differently for men than for women. Specifically, a hazard ratio for interaction significantly greater than one would indicate that digoxin was associated with greater mortality in women than in men. This analysis was then repeated with inclusion of digoxin dose and then serum digoxin level in the model to assess for any evidence that these variables mediated any interaction. Finally, sensitivity analyses were conducted in which baseline blood pressure, BMI, and serum creatinine were included in the analysis, in which the cohort was restricted to participants using concomitant loop diuretics, in which data were included even if the adjusted mortality ratio at the time of collection was not acceptable, and in which subjects were excluded as soon as they crossed over from one exposure group into another.

RESULTS

Characteristics of the Study Population

The study cohort consisted of 17,707 men and 19,227 women who contributed time only without digoxin exposure and 9,487 men and 10,808 women who contributed time with digoxin exposure. Table 1 presents differences in baseline variables between these groups. In general many variables were differently distributed between sexes, as reflected by high standardized differences. However, sex differences were consistent between digoxin exposed and unexposed groups, as reflected by low standardized differences of differences (< 0.1 for all variables).

Baseline drug utilization data were notable for high rates of loop diuretic use (Table 1). Oral anticoagulant use was much more common among digoxin users even after controlling for baseline diagnosis of atrial fibrillation. Examination of trends in drug use during the year after the diagnosis of HF was documented(Figure 1) shows rapid increases in the use of ACE inhibitors, beta-blockers, and spironolactone, with higher rates of use of all of these agents in men compared to women (Figure 1a, Figure 1b).

Blood pressure was recorded for the majority of subjects, while other quantitative covariates such as BMI and serum creatinine were recorded on a minority of subjects. In general rates of missing data were higher for women than for men. Baseline systolic BP mean > 140 was considerably more common in women while a serum creatinine greater that 150 mg/dL was considerably more common in men.

Digoxin Prescribing Patterns Over Time

We found that digoxin use among subjects in this study remained prevalent at about 25% throughout the time period covered by the database (figure 1a and 1b).

Digoxin dose was consistently lower in women than in men (mean daily dose 136 mcg in women, and 159 mcg in men, p<0.001), and the average dose in both groups declined over time by 1.6

mcg per year in men and 1.3 mcg per year in women (p<0.001 for both trends but no significant difference in the rate of decline for men versus women).

Serum digoxin levels were available for 8563 separate measurements covering 4063 individuals. The mean serum level for men was 1.2 ng/dL while for women it was 1.3 ng/dL (p<0.0001 for difference). Linear regression of digoxin level against date yielded an annual decline in mean digoxin level of 0.02 ng/dL per year in men and 0.01 ng/dL per year in women (p<0.0001 for men and p = 0.01 for women, nonsignificant difference in rate between men and women).

Outcomes

Death rates after HF diagnosis without adjustment showed similar rates of mortality in all four cohorts defined by sex and digoxin use (table 2).

Multivariable modeling was pursued in stages (table 3). Briefly, we found that when universally available variables were adjusted for as covariates, there was no evidence of interaction between sex and digoxin use. The adjusted hazard ratio for digoxin exposure in a cohort restricted to women was 1.00 (95% CI 0.96 to 1.06) and for a cohort restricted to men it was identical: 1.00, (95% CI 0.95 to 1.06). A fully adjusted Cox proportional hazards model with a term for the interaction between digoxin use and sex yielded an interaction hazard ratio of 1.02, (95% CI 0.95 to 1.09), where a hazard ratio significantly greater than one would have indicated that digoxin use in women conferred a greater hazard of death than in men.

We examined whether the dose of digoxin mediated any interaction between sex and exposure by including digoxin dose in the model both as an independent variable and as an interaction term with sex, both as a categorical and a continuous variable. The same was done with serum digoxin levels. None of these analyses yielded any significant interaction terms. Low levels of digoxin in the serum (<0.9 ng/dL) were associated with lower mortality than higher levels (HR 0.71, 95% CI 0.67 to0.76, p = 0.007) and this association did not differ by sex.

Sensitivity analyses including restriction of the cohort to persons actively taking loop diuretics, permitting use of data without the adjusted-mortality ratio being up to standard, analyzing the subset of the population with blood pressure, BMI, and serum creatinine available, and excluding persons at the time of crossover to a different exposure category all confirmed the results of the primary analysis, never showing a substantial or statistically significant interaction between sex and digoxin and mortality.

DISCUSSION

The primary finding from this study was the absence of a large interaction between digoxin use and sex affecting mortality, with a 95% confidence interval excluding a hazard ratio > 1.09 for the interaction. This suggests that the association between digoxin and mortality is similar in women and men. Sensitivity analyses did not affect this estimate materially. The other prespecified aims of the study were to assess whether any interaction might be mediated by digoxin dose or by higher serum digoxin levels. Analyses incorporating digoxin dose and serum digoxin levels showed no evidence of such mediation. An interesting incidental finding was rapid increases in the rates of beta-blocker, ACE, and spironolactone use beginning at the same time that major studies were published establishing that these drugs prevented mortality in CHF. [12,13,14] Comparison by sex shows that while overall trends are similar, women consistently have lower levels of use of these mortality-preventing agents compared to men.

A major strength of this study is its large sample size, permitting precise point estimates of effects. The size of the database also permitted numerous sensitivity analyses on sub-populations. The lack of interaction between sex and digoxin use was robust to adjustment for numerous potential confounders and to all sensitivity analyses. The major limitation of this study is that it is non-randomized. Although there is no evidence of substantial confounding of the main study result,

confounding could still bias these results. Access to additional information that was not recorded in THIN, particularly the type (systolic versus diastolic) and severity of heart failure, would have been helpful, but only a randomized trial (ideally a series of such trials in various populations and dosing regimens) could definitively avoid concerns about confounding.

The outcome of this study is discordant with the post-hoc analysis of the DIG trial[2] but consistent with another observational study, done in the SOLVD cohort, of digoxin-sex interaction.[7] It complements that study by having over eight times the sample size and a more diverse cohort. Because the finding of an interaction from the DIG trial was the result of post-hoc analysis it is conceivable that the finding was a type 1 error (false positive), but it is also possible that the DIG trial results were correct and the two observational studies' results are biased by unmeasured confounders that affected the interaction analysis. An interesting incidental finding of this study is that interventions known to reduce mortality in HF are used less in women than in men who carry that diagnosis (Figure 1a, 1b).[12,13,14] It would be premature to conclude that these differences necessarily imply worse care, since there are important differences between the male and female populations with HF that might legitimately affect prescribing practices. However, the systematic differences in drug usage between the sexes deserve further investigation.

In conclusion, this study did not identify a difference between the sexes in the hazard of death associated with the use of digoxin. These results are of use in the context of a clinical question – whether digoxin can be used as safely in women as it can in men – for which there are few randomized data. These results suggest that this drug, with its proven ability to reduce the need for hospitalization, is still a viable therapeutic option in women with heart failure.

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Data sharing: no additional data available.

Table 1. Demographic and Clinical Features of Cohort. MI = Myocardial Infarction; COPD = Chronic Obstructive Pulmonary Disease; BMI = Body Mass Index; Cr = Creatinine; SBP = Systolic Blood Pressure; ACE = Angiotensin Converting Enzyme; ARB = Angiotensin Receptor Blocker; CCB = Calcium Channel Blocker

	Digoxin Non-Use		Standardized Difference	Digoxin Use		Standardized Difference	Standardized Difference of Differences
	Male	Female		Male	Female		
Total (N)	17707	19227		9487	10808		
Baseline Demographics							
Age < 50	3%	2%	0.08	2%	1%	0.11	0.01
Age 50-64	16%	8%	0.25	15%	5%	0.31	0.02
Age 65-74	28%	22%	0.15	30%	19%	0.25	0.07
Age > 74	52%	68%	0.33	54%	75%	0.45	0.08
Baseline Medical History							
History of MI	28%	15%	0.33	22%	12%	0.28	0.05
History of Stroke	14%	13%	0.04	14%	14%	0.00	0.03
History of Diabetes	20%	15%	0.12	16%	13%	0.10	0.02
History of Hypertension	44%	49%	0.10	41%	45%	0.09	0.01
History of COPD	14%	11%	0.11	13%	8%	0.16	0.02
History of Fracture	7%	12%	0.19	6%	11%	0.18	0.01
History of Pneumonia	4%	3%	0.02	4%	3%	0.05	0.02
History of Atrial Fibrillation	10%	8%	0.09	41%	41%	0.00	0.05
Baseline Drug Use							
Statin User	25%	16%	0.23	15%	10%	0.18	0.06
Loop Diuretic User	53%	56%	0.06	59%	58%	0.01	0.05
Aspirin User	42%	32%	0.21	35%	30%	0.11	0.08
Oral Anticoagulant User	9%	5%	0.15	24%	18%	0.13	0.03
Nitrate user	31%	24%	0.15	26%	20%	0.13	0.02
K-Sparing Diuretic User	20%	25%	0.13	24%	29%	0.09	0.02
Spironolactone User	5%	4%	0.04	6%	5%	0.04	0.00
Beta Blocker User	26%	22%	0.10	19%	18%	0.03	0.05
ACE User	36%	28%	0.18	35%	27%	0.18	0.00
ARB User	6%	6%	0.02	5%	5%	0.01	0.02
Thiazide User	16%	23%	0.18	16%	22%	0.16	0.02
CCB User	27%	24%	0.08	23%	21%	0.04	0.03
Lab Data and Vital Signs							
BMI							
% with data	36%	27%	0.20	32%	24%	0.19	0.01

BMI < 18	1%	3%	0.16	1%	4%	0.21	0.05
BMI 18-25	27%	30%	0.07	31%	37%	0.13	0.05
BMI 25-30	41%	31%	0.22	41%	31%	0.21	0.00
BMI 30-35	22%	20%	0.03	19%	16%	0.06	0.02
BMI > 35	10%	16%	0.18	9%	12%	0.09	0.07
Creatinine							
% with data	40%	33%	0.14	33%	29%	0.09	0.04
Cr > 150	16%	8%	0.23	14%	7%	0.23	0.01
Blood Pressure							
% with data	71%	65%	0.12	66%	61%	0.10	0.02
SBP > 140	50%	60%	0.21	47%	58%	0.22	0.01

Table 2. Event rates

	N	Total person-years	Deaths	Deaths Per Person year
Male Digoxin Non-user	17707	53244	4616	0.09
Male Digoxin User	19227	60648	5076	0.08
Female Digoxin Non-user	9487	24804	2324	0.09
Female Digoxin User	10808	30056	2782	0.09

Table 3. Model with all universally available baseline variables. MI = Myocardial Infarction; COPD = Chronic Obstructive Pulmonary Disease; ACE = Angiotensin Converting Enzyme; ARB = Angiotensin Receptor Blocker; CCB = Calcium Channel Blocker

	HR	Lower CI	Upper CI	P
Sex*Digoxin Interaction	1.02	0.95	1.09	0.65
Digoxin Use	1.00	0.95	1.05	0.89
Female Sex	0.86	0.83	0.90	<.0001
Year of CHF Diagnosis	0.99	0.98	0.99	<.0001
Age 50-64	0.42	0.40	0.44	<.0001
Age 65-74	0.58	0.56	0.60	<.0001
Hypertension	0.91	0.88	0.94	<.0001
Stroke	1.34	1.29	1.40	<.0001
MI	1.15	1.11	1.20	<.0001
COPD	1.36	1.30	1.43	<.0001
Fracture	1.08	1.03	1.14	0.001
Diabetes	1.34	1.28	1.40	<.0001
Atrial fibrillation	0.93	0.89	0.97	0.001
Loop diuretic use	1.27	1.23	1.32	<.0001
Potassium-sparing Diuretic use	1.07	1.03	1.11	0.00
Thiazide Diuretic	1.03	0.99	1.07	0.16
Oral Anticoagulant Use	0.92	0.87	0.98	0.001
Beta-blocker use	0.83	0.79	0.86	<.0001
ACE use	0.97	0.93	1.00	0.07
ARB use	0.90	0.82	0.98	0.01
CCB use	0.93	0.90	0.97	0.001

Figure 1a. Trends in Drug Use Among Men

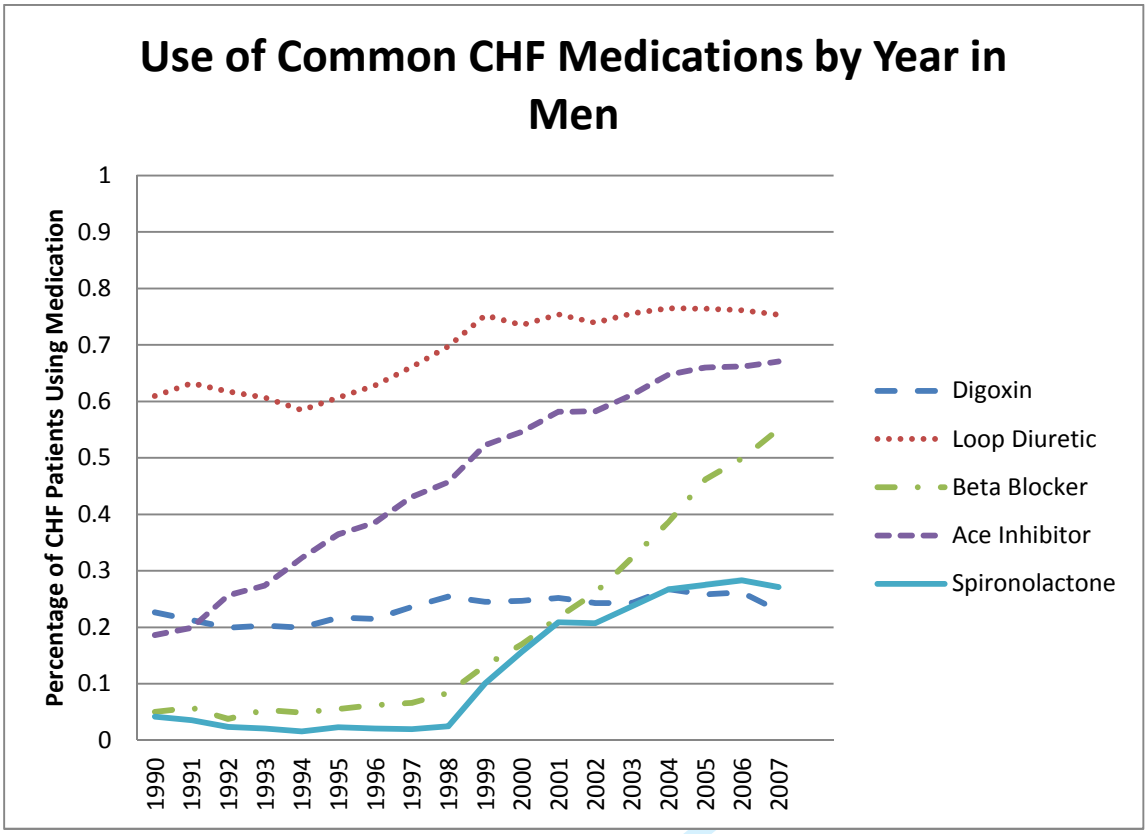
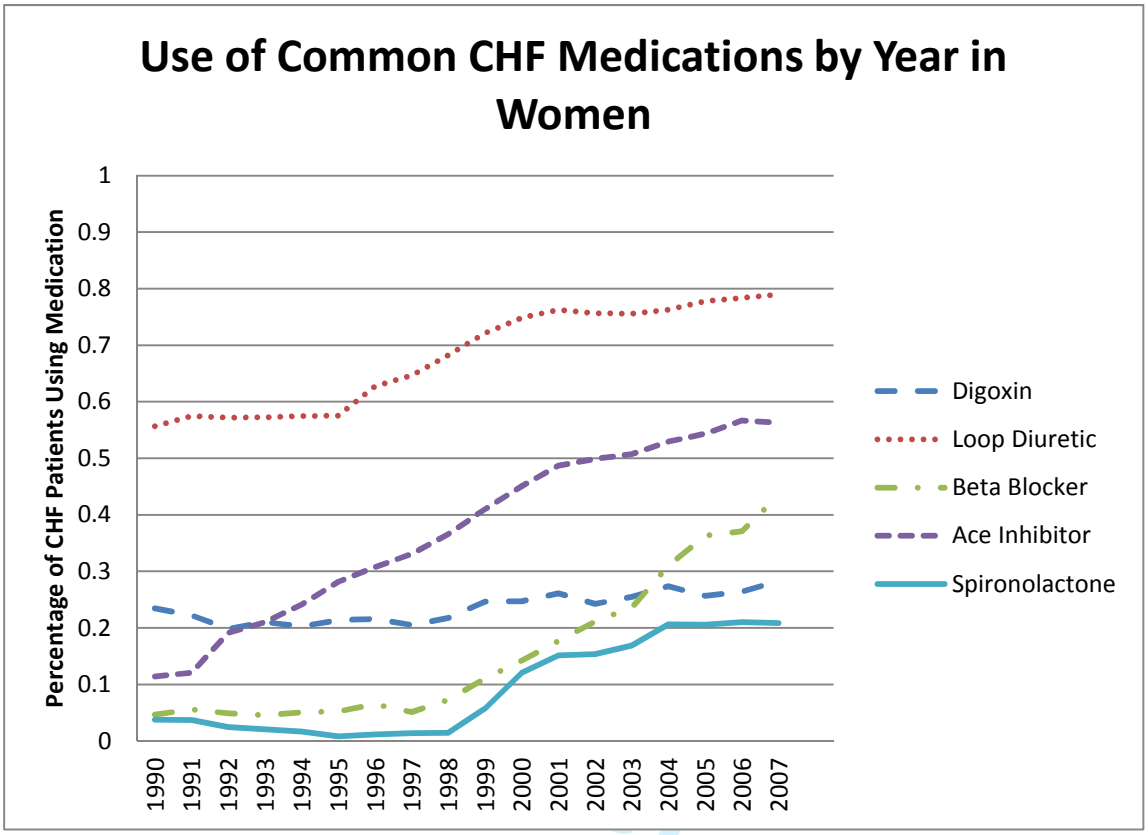


Figure 1b. Trends in Drug Use Among Women



	Item No	Recommendation
Title and abstract		
	1	(*a*) Indicate the study's design with a commonly used term in the title or the abstract **done**
		(*b*) Provide in the abstract an informative and balanced summary of what was done and what was found **done**
Introduction		
Background/rationale	2	Explain the scientific background and rationale for the investigation being reported **done**
Objectives	3	State specific objectives, including any prespecified hypotheses **done**
Methods		
Study design	4	Present key elements of study design early in the paper **done**
Setting	5	Describe the setting, locations, and relevant dates, including periods of recruitment, exposure, follow-up, and data collection **done**
Participants	6	(*a*) *Cohort study*?Give the eligibility criteria, and the sources and methods of selection of participants. Describe methods of follow-up **done**
		(*b*) *Cohort study*?For matched studies, give matching criteria and number of exposed and unexposed **done**
Variables	7	Clearly define all outcomes, exposures, predictors, potential confounders, and effect modifiers. Give diagnostic criteria, if applicable. **done**
Data sources/ measurement	8*	For each variable of interest, give sources of data and details of methods of assessment (measurement). Describe comparability of assessment methods if there is more than one group **done**
Bias	9	Describe any efforts to address potential sources of bias **done**
Study size	10	Explain how the study size was arrived at **done**
Quantitative variables	11	Explain how quantitative variables were handled in the analyses. If applicable, describe which groupings were chosen and why **done**
Statistical methods	12	(*a*) Describe all statistical methods, including those used to control for confounding **done**
		(*b*) Describe any methods used to examine subgroups and interactions **done**
		(*c*) Explain how missing data were addressed **done**

	Item No	Recommendation
		(*d*) *Cohort study*?If applicable, explain how loss to follow-up was addressed **done**
		(*e*) Describe any sensitivity analyses **done**
Results		
Participants	13*	(*a*) Report numbers of individuals at each stage of study?eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed **not applicable**
		(*b*) Give reasons for non-participation at each stage **not applicable**
		(*c*) Consider use of a flow diagram **not applicable**
Descriptive data	14*	(*a*)Give characteristics of study participants (eg demographic, clinical, social) and information on exposures and potential confounders **done**
		(*b*) Indicate number of participants with missing data for each variable of interest **done**
		(*c*) *Cohort study*?Summarise follow-up time (eg average and total amount) **done**
Outcome data	15*	*Cohort study*?Report numbers of outcome events or summary measures over time **done**
Main results	16	(*a*) Report the numbers of individuals at each stage of the study?eg numbers potentially eligible, examined for eligibility, confirmed eligible, included in the study, completing follow-up, and analysed **not applicable**
		(*b*) Give reasons for non-participation at each stage **not applicable**
		(*c*) Consider use of a flow diagram **not applicable**
Other analyses	17	Report other analyses done?eg analyses of subgroups and interactions, and sensitivity analyses **done**
Discussion		
Key results	18	Summarise key results with reference to study objectives **done**
Limitations	19	Discuss limitations of the study, taking into account sources of potential bias or imprecision. Discuss both direction and magnitude of any potential bias **done**
Interpretation	20	Give a cautious overall interpretation of results considering objectives, limitations, multiplicity of analyses, results from similar studies, and other relevant evidence

	Item No	Recommendation
		done
Generalisability	21	Discuss the generalisability (external validity) of the study results **done**
Other information		
Funding	22	Give the source of funding and the role of the funders for the present study and, if applicable, for the original study on which the present article is based **done**

Differential Mortality of Digoxin Users By Gender Over Time: An Observational Study in the THIN Database

James Flory, MD/MSCE candidate (primary author)
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08 November 2008

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I. Specific Aims

Primary Aim 1: test the hypothesis that in patients with heart failure (HF), digoxin use is associated with higher mortality in women but not in men (i.e., that gender is an effect modifier for the association of digoxin use with mortality).

Primary Aim 2: test the hypothesis that effect modification between digoxin use and gender on mortality hazard is attenuated at lower dosage levels (i.e., that there is a three-way effect modification between gender, digoxin, and dose).

Secondary Aim 1: test the hypothesis that digoxin serum levels mediate the gender-digoxin interaction affecting mortality (in the subset of patients for whom these data are available)

II. Background

Several interventions in cardiovascular medicine may have different safety/efficacy profiles depending on gender. For example, digoxin may increase mortality by approximately 20% in women but not in men (1).

The initial signal of this differential effect by gender was identified as part of a post-hoc analysis of the Digitalis Investigation Group (DIG) trial (1). In absolute terms, women assigned to digoxin had a 4.2% higher risk of mortality compared to placebo while men had assigned to digoxin had a 1.6% lower risk of mortality compared to placebo. After multivariable adjustment, the relative risk of mortality in women was statistically significantly increased by digoxin use and the test for an interaction between sex and digoxin affecting mortality was significant at P = 0.014, an effect most likely mediated by higher serum levels in women (2).

The study was accompanied by an editorial which raised three complications in interpreting these results(3). The first was that the finding was post-hoc and could be a spurious false positive. The second letter was that there were significant differences at baseline between men and women (women had higher rates of cardiomegaly, severe heart failure, diabetes, and idiopathic heart failure; men had higher rates of heart failure related to myocardial ischemia and infarction) which could lead to different mortality rates by gender. The third was that, since one of the primary results of DIG had been that digoxin might be associated with increased mortality when serum concentration was greater than 1.0 ng/ml and with reduced mortality when serum concentration was lower than 1.0 ng/ml, it was reasonable to hypothesize that higher serum concentrations in women might be driving the differential effect on mortality by gender. Subsequent letters to the editor of the New England Journal of Medicine elaborated on these points (4, 5, 6). One letter looking at hospital registry data on Sweden found that in clinical practice digoxin serum levels did tend to be higher in female patients and were more often above the therapeutic range, lending credibility to the hypothesis that, if real, the differential mortality might be due to differences in serum level (7).

A different group of investigators subsequently carried out another post hoc analysis of the DIG trial, which examined mortality in women in the control arm versus women in the digoxin arm and stratified by serum digoxin concentration (2). This study found that the hazard ratio for death favored digoxin users (0.8, 95% CI 0.62-1.13) in women with a serum digoxin concentration up to 0.9 ng/ml, while women with a serum concentration of 1.2 ng/ml or greater

had a hazard ratio of 1.33 (95% CI 1.001-1.76) for death. This study further supported the hypothesis that serum digoxin concentration differences between the genders might be driving the sex-digoxin interaction. However, it was not powered to exclude the possibility that gender interaction might be present even after digoxin serum concentration was accounted for.

Finally, an observational study was conducted on patients enrolled in the Studies of Left Ventricular Dysfunction (SOLVD) trial to test for a sex-digoxin interaction affecting mortality, and found no evidence for such an interaction (7). The reason for this disagreement with the DIG post-hoc analysis (1) was not clear, although there were several possible explanations including the fact that the DIG study randomized digoxin use while the SOLVD study did not. Variables including digoxin dose and digoxin serum levels, which might have shed light on the discrepancy, were not included in the analysis. The SOLVD paper ended with the recommendation that further observational studies in other populations be conducted (7).

Further investigation of the potential sex-digoxin (or sex-digoxin-serum-level) interaction is clinically important because digoxin is still widely used by both women and men (8). It remains unclear whether digoxin should be used differently in the different genders (7) and recent publications warn against the use of digoxin in women (9)—an attitude which may unnecessarily curb use of a drug which has been shown to reduce hospitalizations in heart failure. It is plausible that careful maintenance of low digoxin serum concentration would prevent excess mortality in either gender (3), but it is not known whether current clinical practice avoids causing excess mortality in women.

We propose a longitudinal observational study in the THIN database to help answer this question, with prespecified outcomes looking both at gender-digoxin interaction on mortality and at gender-digoxin-dose interaction on mortality. The key advantages which THIN can offer over previously published research include a much larger sample size, use of data from routine clinical practice rather than the artificial environment of a clinical trial, the ability to adjust for digoxin dose within that large sample, and the ability to observe trends over a very long period of use (from the early 1990s up until 2007). Availability of digoxin serum levels on a minority of patients will add further value to the study. If there is a differential effect on mortality by gender, exploration of dose-dependency and serum concentration dependency will give insight into whether there is a way to avoid such effects. Indeed, as the average dose of digoxin has declined over time (8), a longitudinal database may allow a natural experiment to assess whether there was a differential effect by gender in the past which no longer exists under modern dosing practices. Since concerns about digoxin use in women continue to appear in the literature (9), either a positive or a negative finding would help guide clinical decision making, as long as confounding and other forms of bias are acceptably controlled.

III. Research and Methods

A. Overview

Using a retrospective cohort design, we will assess whether the mortality risk for digoxin users interacts with gender. We will also assess whether any interaction depends on the digoxin dose or has changed over the years. The database to be used (THIN, see below) provides

laboratory values on some patients, so it should also be possible to do a subgroup analysis adjusting for serum concentration of digoxin.

The clinical benefit of this study is that it will identify whether digoxin, as it is really being used, is harming women. Exploration of dose-dependency and serum concentration dependency will also give insight into whether there is a way to avoid such effects. Since concerns about digoxin use in women continue to appear in the literature, either a positive or a negative finding will help guide clinical decision making.

B. Rationale for Study Design

The proposed study design is a retrospective cohort study to estimate the hazard-ratio of all-cause deaths in patients chronically taking digoxin versus patients not taking digoxin. A retrospective observational cohort study was chosen for two reasons. First, while a randomized trial provided the initial evidence that digoxin may pose different mortality risks by gender, no randomized set of patients is available with sufficient numbers of outcomes to address all the aims of this protocol. Second, we wanted to study the effects of digoxin on mortality as it has actually been used in ordinary patients over time. This made data collected from routine clinical practice more relevant than data from randomized controlled trial data, provided problems with confounding and bias can be adequately addressed.

Confounding will be an issue in the design of this study, but the problem of confounding will be attenuated in two ways. The most problematic comparison would be to assess the direct effect of digoxin on mortality, since it is quite likely that patients who receive digoxin differ systematically from those who do not. However, the first primary outcome of this study is an assessment of drug-gender interaction. For this comparison to be confounded, a baseline difference between digoxin users and non-users would have to also differ by gender, a less likely scenario. Furthermore, the second primary outcome of this study is an assessment of drug-gender-dose interaction. Changes in dosing practices for digoxin are unlikely to be influenced strongly by gender; indeed, there is a strong secular trend towards lower doses of digoxin (8), which suggests that much variation in dosing practices is driven by factors other than patient characteristics.

C. Source Population/Database Description

The research design for this study is a retrospective cohort study. The source of the cohort will be a database from the United Kingdom called THIN (The Health Improvement Network). This database contains over 5 million individuals and has been validated for a variety of outcomes (10). The analysis will be restricted to individuals with HF, with digoxin as the exposure of interest, and assess hazard ratios for several relevant outcomes (all cause mortality as the primary outcome, with sudden cardiac death, digoxin toxicity, and hospitalization as secondary outcomes). THIN provides lab values on a subset of patients, so it should also be possible to execute a subgroup analysis adjusting for actual serum concentration of digoxin.

At the Center for Clinical Epidemiology and Biostatistics (CCEB), THIN can be used at minimal cost. A random 10% sample of the whole database is available for pilot work and can be used immediately for refinement of power calculations and development of analytic code for this

project. The primary author is familiar with the software packages (particularly MySQL and SAS) needed for this and will write the analytic code. The SAS package in particular includes procedures for the COX proportional hazard regression that will be used in the primary analysis (see next section).

D. Subjects

The analysis will be restricted to individuals with heart failure (HF). As detailed below, subjects must be treated with medication for this chronic disease.

1. Exposed Population

Digoxin exposure will be defined as consecutive, recurrent prescriptions (2 or more) of digoxin. Individuals will contribute exposed person-time from 90 days after their first prescription of digoxin until 30 days after their last prescription.

Individuals will contribute exposed person-time from the date of their first prescription of digoxin until 30 days after their last prescription. Exposure for this study is meant to capture current use of digoxin.

A last prescription is defined as a prescription not followed by another within 90 days. To clarify, if somebody takes digoxin for many years but during that time has a single gap of greater than 90 days, they will contribute two blocks of exposed time.

Of note, individuals in the control population are required to be on other medications associated with HF in order to contribute followup time. A parallel requirement is not in place for the cases (who contribute person-time even if digoxin is their only medication) in the primary analysis. There is a possibility that this would create bias by allowing more individuals with less severe or symptomatic HF into the case arm. However, in a secondary analysis, cases will be restricted to those with another medication in addition to digoxin.

At least one diagnostic code for heart failure will be required for any case.

2. Control Population

The control population will consist of individuals with HF who are being treated with medication for that condition. Only person time during such medical treatment (or for 30 days after the last prescription) will be included in the study. The reason for this rule is that a diagnostic code for HF unaccompanied by any medical treatment is likely to reflect either a mistaken code; an extremely mild case, or a patient so ill that they cannot adhere to chronic outpatient medication. Chronic medication use also implies frequent followup with a physician such that morbidities and mortality will be appropriately recorded. Medications commonly used chronically for heart failure include beta-blockers, diuretics (including aldosterone antagonists for severe cases), and ACE inhibitors.

Because all of these medications have other indications in addition to heart failure, at least one diagnostic code for heart failure will be required for any control. The diagnostic code must appear within 6 months of initiation of medication, to minimize ambiguous situations in which use of a medication like a beta-blocker may precede the onset of heart failure.

3. Exclusion Criteria

The only exclusion criterion for this study is that patients must have one year or more of follow-up time in the database prior to their incident diagnosis.

E. Covariates

Covariates in the analysis will include calendar year, age, sex, BMI and smoking status, (39% missing data, so BMI and smoking will only be included in a sensitivity analysis), history of hypertension, history of MI, history of stroke, history of hospitalization, and history of diabetes. Baseline statin use will also be used as a covariate. The most useful covariates (ie, consistently available and associated with the outcome) are likely to be medication use. The cornerstones of HF pharmacotherapy are beta-blockers, ACE inhibitors, and diuretics. These drugs will be entered as covariates and, numbers permitting, cases and controls will be matched by medication regimen. Diagnostic codes defining ejection fraction, severity of HF, or type of HF (systolic versus diastolic) are unlikely to be available on enough patients to be used in any but secondary analyses. Please see code appendix for additional details.

Baseline covariates will be assessed in the year prior to the index date for the study. For both cases and controls, the index date is the date of initiation of medical treatment for HF. Medication exposure at baseline consist of the constellation of medications intiated before or within 90 days of the beginning of treatment for HF. Medication exposure will also be analysed as a time-varying covariate as a sensitivity analysis; if this sensitivity analysis alters the study conclusions then more sophisticated methods for analyzing time varying covariates, such as a marginal structural model, will be employed.

F. Outcome

The primary outcome for this study is death, which is recorded in the THIN database as a death date. Secondary outcomes, such as death from cardiac arrhythmia and digoxin toxicity, will be defined using medical diagnosis codes (see code appendix).

G. Codes

Medical and drug codes were identified through text searches of the THIN medical codes databases, hand screening, and consultation with other researchers at the University of Pennsylvania who have done research on digoxin and on HF. The codes are included as an appendix.

H. Analysis

The initial analysis will be descriptive, comparing exposed and unexposed individuals. Differences between groups will be tested for significance using chi-square and t-tests depending on whether the variable of interest is dichotomous or continuous. Incidence rates in each group will be calculated, then hazard ratios will be calculated using Cox proportional hazards models. Both unadjusted and adjusted models will be run. Adjusted models will include all variables outlined in the Covariate section above.

The final analysis to address the first primary aim will most likely be presented as two multivariable Cox models, one for males and one for females. The formal significance test for a digoxin-gender interaction, however, will most easily be done via a single Cox model which includes sex, digoxin exposure, and a sex-digoxin exposure interaction term in the model.

To address the second primary aim, average daily dose of digoxin will be calculated and the analysis will be stratified by quartiles of average daily dose. Because of the strong secular downward trend in digoxin dose with time, it will be particularly important to control for calendar year in this analysis. In an exploratory component of this aim, we will investigate whether calendar year, possibly in combination with physician prescribing preference, can be used as an instrumental variable and whether this adjustment changes the results of the analysis. In theory, an instrumental variable can control for confounding that is not necessarily captured by conventional multivariable adjustment (11).

To address the third (and secondary) aim, we will identify patients for whom digoxin serum levels are available in the database. For that subset of patients, the analyses described for aim 1 can be repeated but including serum level of digoxin in the model. It is possible that there will be insufficient data of this kind for us to complete this aim.

In all aims, sensitivity analysis for the potential effect of unmeasured confounding will also be conducted (12).

I. Sample Size

Pilot data indicate that approximately 800,000 patients in the THIN database have had multiple prescriptions for digoxin at some point and that 30,000 of these patients have died. While these pilot numbers do not incorporate exclusion criteria or assess how many of these deaths occurred while the patient was taking digoxin, they suggest that the number of eligible outcomes is likely to be large and will certainly support aims 1 and 2. Even if only 1% of these deaths are eligible outcomes in digoxin-exposed individuals, the study should be 90% powered at an alpha of 0.05 to detect a hazard ratio for the digoxin-gender interaction of 1.3 (analysis done using Schoenfeld's equations for time-to-event analysis).

J. Plan for reporting and following up results

This project is intended for peer-reviewed publication. The primary parameter estimates from this research will be a hazard ratio for a digoxin-gender interaction (primary aim 1, regarding the basic question of whether such an interaction is present). The results of primary aim 2 will be more complicated to present but we anticipate reporting multiple digoxin-gender interaction hazard ratios after stratification by calendar year.

K. Anticipated Limitations

●Representativeness of Cohort: Since exposed and unexposed are drawn from a large population sample and treated as a cohort, they should be representative of the UK population.

●Misclassification of Exposure: The exposure is use of a prescription drug. Prescriptions issued are automatically captured by the systems that contribute data to THIN and these data are considered highly reliable. As a proxy for use of the drug, this is not a perfectly valid measure because patients may never fill or take the prescription. However, such misclassification should be minimal and nondifferential. Dosing levels will be computed from prescription data as has been done in other studies (8); similar concerns about nondifferential misclassification apply.

●Misclassification of Outcome:

○Mortality is recorded in THIN as a mortality code. The specificity of this measure should be good and can be confirmed by checking to make sure that medical followup ends at the time of reported death. Sensitivity may be imperfect if some deaths are not recorded and merely appear as the end of followup in THIN; however, this is unlikely to be a major problem and would most likely result in nondifferential bias to the null.

●Confounding by Indication: confounding by indication is likely to be a less severe problem because this study focuses on interactions rather than on the main drug effect. Adjustment for covariates will also be employed.

IV. Implications

This project has implications for basic pharmacologic science, clinical practice, epidemiological methods, and future research.

A. Basic Science

The major basic science contribution of this project would be to assess whether there is a dose-gender-digoxin three-way interaction. A dose-response relationship, which is unlikely to be due to confounding, would support the current hypothesis that differential mortality is due to the same dosing level resulting in different serum concentration in the different genders.

B. Clinical Practice

This project is focused on clinical practice: the question is whether there is a need for different usage guidelines for digoxin based on gender. In particular, primary aim 2 will address the hypothesis that modern dosing practices equalize the risk between genders.

C. Epidemiological Methods

This project will result in careful examination of two methodological questions. The first is whether interactions are typically less confounded than direct effects. The second is whether a strong secular trend in how a drug is used can lead to a convincing demonstration of how the changing practice impacts outcomes.

V. Timeline and Logistics

A. Human Subjects Protections

1. Approval from Penn IRB
2. Approval from ethics bodies governing THIN

B. Grant Support

For support for this study, I will apply to the CTSA-ACARD Internal Small Grant Program (attached). It is possible to apply for any of 5 levels of support in working with the THIN database. For this project, the most appropriate is level 4.

Level 4 support includes exporting data files from THIN to a PC environment. ACARD intends to fund small grants to export analytic data files from the full THIN (residing on a server) for use on a PC. This funding is for researchers who have pilot-tested their variable creation, cohort selection criteria, and statistical code using the 10% sample data residing on a dedicated PC (known as 'Victoria'). These grants are for $1000 each, all of which must be used for the cost of the Biostatistical Analysis Center (BAC) to export the data from the full THIN database for further analysis by the investigators. The BAC will implement the SQL, SAS, and/or STATA code provided by the applicant to create the study data files. The BAC will not be responsible for determining the integrity of the code. If selected for funding, the funds will be directly transferred from ACARD to BAC.

I am writing my own SQL, SAS, and STATA code to conduct this analysis with the guidance of my thesis advisor and my biostatistics advisor. PC resources available within the CCEB will be sufficient to implement this code.

C. Timeline

To do	*Sep. '08*	*Oct.*	*Nov. '08*	*Dec.*	*Jan.*	*Feb.*
Develop protocol	**********					
Penn IRB approval (done)		********				
UK Ethics Approval			**********			
Write, pilot, and debug SAS/SQL code			**********	**********	**********	
Data extraction and						*

analysis	
Writeup primary results	*****

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Observational Cohort Study of the Safety of Digoxin Use in Women with Heart Failure

Journal:	*BMJ Open*
Manuscript ID:	bmjopen-2012-000888.R1
Article Type:	Research
Date Submitted by the Author:	08-Mar-2012
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Primary Subject Heading:	Cardiovascular medicine
Secondary Subject Heading:	Epidemiology, Evidence based practice, Pharmacology and therapeutics
Keywords:	Cardiac Epidemiology < CARDIOLOGY, Heart failure < CARDIOLOGY, EPIDEMIOLOGY

Observational Cohort Study of the Safety of Digoxin Use in Women with Heart Failure

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Keywords: heart failure, digoxin, mortality, sex

Support: *FOCUS Medical Student Fellowship in Women's Health* supported by the Edna G. Kynett Memorial Foundation, Pharmacoepidemiology Training Funds donated to the Center for Clinical Epidemiology and Biostatistics of the University of Pennsylvania School of Medicine. The study sponsors had no influence over design of the study, the handling of data, the composition of the manuscript, or the decision to submit it.

All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author). SH has consulted for Abbott Laboratories. BLS has consulted for Abbott Laboratories, received payment for lectures from Teva Pharmaceutical Industries, and has consulted for Teva and for GlaxoSmithKline. All authors on this manuscript have a connection to a training program which has received funding from Abbott Laboratories and GlaxoSmithKline. None of these connections related directly to digoxin. These companies are among the over 50 corporations that manufacture or distribute digoxin.

Guarantorship and Contributorship: JHF is the guarantor of this manuscript, having been primarily responsible for study design, data analysis including programming and biostatistics, and manuscript composition. JHF, BK, KH, SB, JM, CR, BLS , and SH all made substantial contributions to the conception and design of this study. JHF was primarily responsible for analysis and interpretation of the data but all listed authors made significant contributions to this as well. All of the listed authors made substantial contributions in drafting the article or revising it critically for important intellectual content.

SUMMARY

Objectives This study aims to assess whether digoxin has a different effect on mortality risk for women than it does for men in patients with heart failure (HF).

Design This study uses the United Kingdom-based The Health Information Network (THIN) population database in a cohort study of the impact of digoxin exposure on mortality for men and women who carry the diagnosis of HF. Digoxin exposure was assessed based on prescribing data. Multivariable Cox proportional hazards models were used to assess whether there was an interaction between sex and digoxin affecting mortality hazard.

Setting The setting was primary care outpatient practices.

Participants The study cohort consisted of 17,707 men and 19,227 women with the diagnosis of HF who contributed only time without digoxin exposure and 9,487 men and 10,808 women with the diagnosis of HF who contributed time with digoxin exposure.

Main Outcome Measures The main outcome measure was all-cause mortality.

Results The primary outcome of this study was the absence of a large interaction between digoxin use and sex affecting mortality. For men digoxin use was associated with a hazard ratio for mortality of 1.00, while for women the hazard ratio was also 1.00 (p-value for interaction 0.65). SThe results of sensitivity analyss were consistent with those of the primary analysis.

Conclusion Observational data do not support the concern that there is a substantial increased risk of mortality due to the use of digoxin in women. This finding is consistent with previous observational studies but discordant with results from a post-hoc analysis of a randomized controlled trial of digoxin versus placebo.

INTRODUCTION

It has been hypothesized that digoxin, when used in the treatment of heart failure (HF), may increase mortality by approximately 20% in women but not in men. This hypothesis is based on a post-hoc analysis in 6800 patients by Rathore and colleagues of the Digitalis Investigation Group (DIG) trial, a placebo-controlled randomized trial that showed digoxin did not affect overall mortality but did reduce hospitalizations in patients with HF.[1] The post-hoc analysis examined mortality effects by sex and found that compared to placebo digoxin conferred reduced mortality in men (absolute difference -1.6%, 95% CI -4.2% to 1.0%) and increased mortality in women (absolute difference 4.2%, 95% CI -0.5% to 8.8%), with a statistically significant interaction (p value= 0.034).[2] One proposed mechanism is that women may have higher mean serum digoxin levels than men. There is evidence that even within the therapeutic range higher serum digoxin levels are associated with increased mortality, and experts have argued that maintenance of serum digoxin levels at the low end of the therapeutic range may be the key to safe, effective use of the drug in either sex.[3-7]

Because the sex-digoxin interaction was based on post-hoc analyses, it needs cautious interpretation. Yet, if true, the finding is clinically important because digoxin continues to be widely used by both women and men.[8] It remains unclear whether digoxin should be used differently in the different sexes, and concerns about its use in women continue to appear in the literature.[9,10] Unwarranted recommendations against use of digoxin in women would deprive a large population of a medicine with demonstrated ability to prevent hospitalizations.

Further randomized trials evaluating the interaction between digoxin and sex have not emerged. One observational study based on the Studies of Left Ventricular Dysfunction (SOLVD) cohort found no difference in digoxin's effect on mortality between sexes, but concluded that additional research in other populations is still needed.[10] Limitations of that study included restriction to a population with severely reduced ejection fraction (<35%) and relatively small sample size (n=6797). Another observational study performed on 2841 propensity-score-matched patients with heart failure also did not report any difference in digoxin's effect on mortality between the sexes; this study was primarily limited by a relatively small size and restriction to a single clinical center.[11] We sought to conduct a much larger study in a broader population that could help assess whether digoxin increases mortality in women compared to men.

METHODS

Study Design: We conducted a retrospective cohort study to test the primary null hypothesis that sex was not an effect modifier for the association of digoxin use with mortality. We also conducted planned secondary analyses of whether effect modification was mediated by digoxin dose or by serum digoxin concentration. This study was approved by the Institutional Review Board of the University of Pennsylvania. Analyses were conducted using SAS 9.1.

Study Population: The study used the Health Improvement Network (THIN) database, a primary care electronic medical record database in the UK. This database contains over 5 million individuals who have contributed person-time from over 300 different general practices from 1986 to 2008. THIN includes demographic information on patients, as well as records of prescribed drugs, medical diagnoses (coded as READ codes), as well as vital signs and laboratory values on a subset of patients. It is a representative subset of the United Kingdom's general population.[12]

Cohort Definition: The analysis was restricted to individuals with a diagnostic code for HF. Once that condition was met, patients could contribute follow-up time as long as they were being given regular prescriptions of at least one drug consistent with the treatment for HF (as described below). Information was only used if it was collected during times when the adjusted mortality ratio, a quality control measure used in THIN to identify practices which are recording deaths accurately, was within acceptable standards for a given practice.[13] It was possible for one individual to contribute both exposed and unexposed time. The result was four cohorts with some within-sex crossover: men on digoxin, men not on digoxin, women on digoxin, women not on digoxin.

Exposed Group: Exposed subjects were defined as individuals who carried the diagnosis of HF and were receiving two or more consecutive prescriptions for digoxin. Individuals were assumed to be on digoxin from the date of receipt of a prescription until 30 days after their prescription ran out (the intended duration of a prescription was either computed from information on daily dose and number of tablets, or imputed as 30 days if that information was not available).

Unexposed Group: Unexposed subjects were defined as individuals who carried a diagnosis of HF and were receiving two or more consecutive prescriptions for a loop diuretic, beta blocker, angiotensin converting enzyme (ACE) inhibitor, or angiotensin receptor blocker (ARB). They only contributed person-time to the cohort while receiving these drugs; they were assumed to be on a drug from the date of receipt of a prescription until 30 days after their prescription ran out, which was defined with the same criteria used for digoxin exposure. These are drugs that, while not specific for HF, are commonly used for that condition.

Definition of Outcome: The outcome for this study was all-cause mortality, as documented by a date of death in the THIN demographics file.

Definition of Covariates: Baseline covariates consisted of diagnostic codes for common comorbidities of HF, including hypertension, stroke, myocardial infarction (MI), chronic obstructive pulmonary disease (COPD), diabetes, and atrial fibrillation. We also assessed age, sex, and baseline use of drugs commonly used in HF, notably aspirin, statins, oral anticoagulants, potassium-sparing diuretics, and the HF-associated drugs listed above. To be included as a baseline variable drug use had to be in the year prior to HF diagnosis; medical history such as a history of hypertension could be coded at any time prior to HF diagnosis. Blood pressure, body mass index (BMI), and a variety of lab values including serum creatinine were available on a minority of participants and were used as baseline variables in sensitivity analysis. Once a patient began to contribute exposure-time, baseline variables were no longer updated.

Statistical Analysis: Because of the large sample size, we anticipated that even clinically insignificant differences between baseline variables could be statistically significant and therefore described baseline variables using standardized differences.[14] Standardized differences are a balance diagnostic used to assess how similar groups are at baseline – conceptually they are equal to the difference of a variable's mean between two groups divided by the standard deviation for that variable. A standardized difference of < 0.1, by convention, is considered to indicate good balance of groups on that variable.[14] For categorical variables a standardized difference between men and women was calculated as

$\frac{p_{women} - p_{men}}{\sqrt{(\frac{p_{women}(1-p_{women}) + p_{men}(1-p_{men})}{2})}}$, where p = proportion of the population falling into a category. In addition, because the most relevant question was whether these between-sex differences were consistent in both digoxin-exposed and digoxin-unexposed groups, this equation was extended to treat the inter-sex differences for the exposed and unexposed groups as variables which in turn had their own standardized differences:

$$[(p_{women,exposed} - p_{men,exposed}) - (p_{women,unexposed} - p_{men,unexposed})]/\sqrt{[(p_{women,exposed}(1-p_{women,exposed}) + p_{men,exposed}(1-p_{men,exposed}) + p_{women,unexposed}(1-p_{women,unexposed}) + p_{men,unexposed}(1-p_{men,unexposed}))/4]}$$

Total follow-up time for all individuals was assessed, with digoxin use, digoxin dose, and serum digoxin concentrations as time-varying covariates in secondary analysis. In the primary analysis digoxin dose and serum concentration were ignored. A Cox proportional hazards model was used to calculate hazard ratios for sex, digoxin use, and the interaction of sex and digoxin use, controlling for the covariates specified above.[15]

The primary parameter of interest was the adjusted hazard ratio for the interaction of sex and digoxin use, where a significant deviation from the null would indicate that digoxin use was associated with mortality differently for men than for women. Specifically, a hazard ratio for interaction significantly greater than one would indicate that digoxin was associated with greater mortality in women than in men. This analysis was then repeated with inclusion of digoxin dose and then serum digoxin level in the model to assess for any evidence that these variables mediated any interaction. Finally, sensitivity analyses were conducted in which baseline blood pressure, BMI, and serum creatinine were included in the analysis, in which the cohort was restricted to participants using concomitant loop diuretics, in which comorbid conditions were defined based only on data from the one year preceding cohort entry, in which patients with a diagnosis of atrial fibrillation were excluded, in which data were included even if the adjusted mortality ratio at the time of collection was not acceptable, and in which subjects were excluded as soon as they crossed over from one exposure group into another.

RESULTS

Characteristics of the Study Population

The study cohort consisted of 17,707 men and 19,227 women who contributed time only without digoxin exposure and 9,487 men and 10,808 women who contributed time with digoxin exposure. Table 1 presents differences in baseline variables between these groups. In general many variables were differently distributed between sexes, as reflected by high standardized differences. However, sex differences were consistent between digoxin exposed and unexposed groups, as reflected by low standardized differences of differences (< 0.1 for all variables).

Baseline drug utilization data were notable for high rates of loop diuretic use (Table 1). Oral anticoagulant use was much more common among digoxin users even after controlling for baseline diagnosis of atrial fibrillation. Examination of trends in drug use during the year after the diagnosis of HF was documented(Figure 1) shows rapid increases in the use of ACE inhibitors, beta-blockers, and spironolactone, with higher rates of use of all of these agents in men compared to women (Figure 1a, Figure 1b).

Blood pressure was recorded for the majority of subjects, while other quantitative covariates such as BMI and serum creatinine were recorded on a minority of subjects. In general rates of missing data were higher for women than for men. Baseline systolic BP mean > 140 was considerably more

common in women while a serum creatinine greater that 150 mg/dL was considerably more common in men.

Digoxin Prescribing Patterns Over Time

We found that digoxin use among subjects in this study remained prevalent at about 25% throughout the time period covered by the database (figure 1a and 1b).

Digoxin dose was consistently lower in women than in men (mean daily dose 136 mcg in women, and 159 mcg in men, p<0.001), and the average dose in both groups declined over time by 1.6 mcg per year in men and 1.3 mcg per year in women (p<0.001 for both trends but no significant difference in the rate of decline for men versus women).

Serum digoxin levels were available for 8563 separate measurements covering 4063 individuals. The mean serum level for men was 1.2 ng/dL while for women it was 1.3 ng/dL (p<0.0001 for difference). Linear regression of digoxin level against date yielded an annual decline in mean digoxin level of 0.02 ng/dL per year in men and 0.01 ng/dL per year in women (p<0.0001 for men and p = 0.01 for women, nonsignificant difference in rate between men and women).

Outcomes

Death rates after HF diagnosis without adjustment showed similar rates of mortality in all four cohorts defined by sex and digoxin use (table 2).

Multivariable modeling was pursued in stages (table 3). Briefly, we found that when universally available variables were adjusted for as covariates, there was no evidence of interaction between sex and digoxin use. The adjusted hazard ratio for digoxin exposure in a cohort restricted to women was 1.00 (95% CI 0.96 to 1.06) and for a cohort restricted to men it was identical: 1.00, (95% CI 0.95 to 1.06). A fully adjusted Cox proportional hazards model with a term for the interaction between digoxin use and sex yielded an interaction hazard ratio of 1.02, (95% CI 0.95 to 1.09), where a hazard ratio significantly greater than one would have indicated that digoxin use in women conferred a greater hazard of death than in men.

We examined whether the dose of digoxin mediated any interaction between sex and exposure by including digoxin dose in the model both as an independent variable and as an interaction term with sex, both as a categorical and a continuous variable. The same was done with serum digoxin levels. None of these analyses yielded any significant interaction terms. Low levels of digoxin in the serum (<0.9 ng/dL) were associated with lower mortality than higher levels (HR 0.71, 95% CI 0.67 to 0.76, p = 0.007) and this association did not differ by sex.

Sensitivity analyses including restriction of the cohort to persons actively taking loop diuretics, permitting use of data without the adjusted-mortality ratio being up to standard, analyzing the subset of the population with blood pressure, BMI, and serum creatinine available, defining comorbid conditions based only on data from the one year preceding cohort entry, excluding patients with a diagnosis of atrial fibrillation, and excluding persons at the time of crossover to a different exposure category all confirmed the results of the primary analysis, never showing a substantial or statistically significant interaction between sex and digoxin and mortality.

DISCUSSION

The primary finding from this study was the absence of a large interaction between digoxin use and sex affecting mortality, with a 95% confidence interval excluding a hazard ratio > 1.09 for the interaction. This suggests that the association between digoxin and mortality is similar in women and men. Sensitivity analyses did not affect this estimate materially.

The other prespecified aims of the study were to assess whether any interaction might be mediated by digoxin dose or by higher serum digoxin levels, both of which are thought to potentially

affect mortality rates.[3-7] Analyses incorporating digoxin dose and serum digoxin levels showed no evidence of such mediation. We did note an association between lower serum digoxin levels and lower mortality, but since such an association could easily be strongly confounded (for example, if more symptomatic patients were pushed to higher serum digoxin levels), this paper does not directly support or refute the claim that lower serum levels of digoxin lead to lower mortality risk.

An interesting incidental finding was rapid increases in the rates of beta-blocker, ACE, and spironolactone use beginning at the same time that major studies were published establishing that these drugs prevented mortality in CHF.[16-18] Comparison by sex shows that while overall trends are similar, women consistently have lower levels of use of these life-saving agents compared to men.

A major strength of this study is its large sample size, permitting precise point estimates of associations. The size of the database also permitted numerous sensitivity analyses on sub-populations. The lack of interaction between sex and digoxin use was robust to adjustment for numerous potential confounders and to all sensitivity analyses. The major limitation of this study is that it is non-randomized. Although there is no evidence of substantial confounding of the main study result, confounding could still bias these results. Access to additional information that was not recorded in THIN, particularly the type (systolic versus diastolic) and severity of heart failure, would have been helpful, but only a randomized trial (ideally a series of such trials in various populations and dosing regimens) could definitively avoid concerns about confounding.

The outcome of this study is discordant with the post-hoc analysis of the DIG trial[2] but consistent with two prior observational studies which did not observe any digoxin-sex interaction.[10, 11] It complements those prior studies by having over eight times their sample sizes and a more diverse cohort. Because the finding of an interaction from the DIG trial was the result of post-hoc analysis it is conceivable that the finding was a type 1 error (false positive), but it is also possible that the DIG trial results were correct and the observational results are biased by unmeasured confounders that affected the interaction analysis. An interesting incidental finding of this study is that interventions known to reduce mortality in HF are used less in women than in men who carry that diagnosis (Figure 1a, 1b).[16-18] It would be premature to conclude that these differences necessarily imply worse care, since there are important differences between the male and female populations with HF that might legitimately affect prescribing practices. However, the systematic differences in drug usage between the sexes deserve further investigation.

In conclusion, this study did not identify a difference between the sexes in the hazard of death associated with the use of digoxin. These results are of use in the context of a clinical question – whether digoxin can be used as safely in women as it can in men – for which there are few randomized data. These results suggest that this drug, with its proven ability to reduce the need for hospitalization, is still a viable therapeutic option in women with heart failure.

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Data sharing: no additional data available.

Table 1. Demographic and Clinical Features of Cohort. MI = Myocardial Infarction; COPD = Chronic Obstructive Pulmonary Disease; BMI = Body Mass Index; Cr = Creatinine; SBP = Systolic Blood Pressure; ACE = Angiotensin Converting Enzyme; ARB = Angiotensin Receptor Blocker; CCB = Calcium Channel Blocker

	Digoxin Non-Use		Standardized Difference	Digoxin Use		Standardized Difference	Standardized Difference of Differences
	Male	Female		Male	Female		
Total (N)	17707	19227		9487	10808		
Baseline Demographics							
Age < 50	3%	2%	0.08	2%	1%	0.11	0.01
Age 50-64	16%	8%	0.25	15%	5%	0.31	0.02
Age 65-74	28%	22%	0.15	30%	19%	0.25	0.07
Age > 74	52%	68%	0.33	54%	75%	0.45	0.08
Baseline Medical History							
History of MI	28%	15%	0.33	22%	12%	0.28	0.05
History of Stroke	14%	13%	0.04	14%	14%	0.00	0.03
History of Diabetes	20%	15%	0.12	16%	13%	0.10	0.02
History of Hypertension	44%	49%	0.10	41%	45%	0.09	0.01
History of COPD	14%	11%	0.11	13%	8%	0.16	0.02
History of Fracture	7%	12%	0.19	6%	11%	0.18	0.01
History of Pneumonia	4%	3%	0.02	4%	3%	0.05	0.02
History of Atrial Fibrillation	10%	8%	0.09	41%	41%	0.00	0.05
Baseline Drug Use							
Statin User	25%	16%	0.23	15%	10%	0.18	0.06
Loop Diuretic User	53%	56%	0.06	59%	58%	0.01	0.05
Aspirin User	42%	32%	0.21	35%	30%	0.11	0.08
Oral Anticoagulant User	9%	5%	0.15	24%	18%	0.13	0.03
Nitrate user	31%	24%	0.15	26%	20%	0.13	0.02
K-Sparing Diuretic User	20%	25%	0.13	24%	29%	0.09	0.02
Spironolactone User	5%	4%	0.04	6%	5%	0.04	0.00
Beta Blocker User	26%	22%	0.10	19%	18%	0.03	0.05
ACE User	36%	28%	0.18	35%	27%	0.18	0.00
ARB User	6%	6%	0.02	5%	5%	0.01	0.02
Thiazide User	16%	23%	0.18	16%	22%	0.16	0.02
CCB User	27%	24%	0.08	23%	21%	0.04	0.03
Lab Data and Vital Signs							
BMI							
% with data	36%	27%	0.20	32%	24%	0.19	0.01

BMI < 18	1%	3%	0.16	1%	4%	0.21	0.05
BMI 18-25	27%	30%	0.07	31%	37%	0.13	0.05
BMI 25-30	41%	31%	0.22	41%	31%	0.21	0.00
BMI 30-35	22%	20%	0.03	19%	16%	0.06	0.02
BMI > 35	10%	16%	0.18	9%	12%	0.09	0.07
Creatinine							
% with data	40%	33%	0.14	33%	29%	0.09	0.04
Cr > 150	16%	8%	0.23	14%	7%	0.23	0.01
Blood Pressure							
% with data	71%	65%	0.12	66%	61%	0.10	0.02
SBP > 140	50%	60%	0.21	47%	58%	0.22	0.01

Table 2. Event rates

	N	Total person-years	Deaths	Deaths Per Person year
Male Digoxin Non-user	17707	53244	4616	0.09
Male Digoxin User	19227	60648	5076	0.08
Female Digoxin Non-user	9487	24804	2324	0.09
Female Digoxin User	10808	30056	2782	0.09

Table 3. Model with all universally available baseline variables. MI = Myocardial Infarction; COPD = Chronic Obstructive Pulmonary Disease; ACE = Angiotensin Converting Enzyme; ARB = Angiotensin Receptor Blocker; CCB = Calcium Channel Blocker

	HR	Lower CI	Upper CI	P
Sex*Digoxin Interaction	1.02	0.95	1.09	0.65
Digoxin Use	1.00	0.95	1.05	0.89
Female Sex	0.86	0.83	0.90	<.0001
Year of CHF Diagnosis	0.99	0.98	0.99	<.0001
Age 50-64	0.42	0.40	0.44	<.0001
Age 65-74	0.58	0.56	0.60	<.0001
Hypertension	0.91	0.88	0.94	<.0001
Stroke	1.34	1.29	1.40	<.0001
MI	1.15	1.11	1.20	<.0001
COPD	1.36	1.30	1.43	<.0001
Fracture	1.08	1.03	1.14	0.001
Diabetes	1.34	1.28	1.40	<.0001
Atrial fibrillation	0.93	0.89	0.97	0.001
Loop diuretic use	1.27	1.23	1.32	<.0001
Potassium-sparing Diuretic use	1.07	1.03	1.11	0.00
Thiazide Diuretic	1.03	0.99	1.07	0.16
Oral Anticoagulant Use	0.92	0.87	0.98	0.001
Beta-blocker use	0.83	0.79	0.86	<.0001
ACE use	0.97	0.93	1.00	0.07
ARB use	0.90	0.82	0.98	0.01
CCB use	0.93	0.90	0.97	0.001

Differential Mortality of Digoxin Users By Gender Over Time: An Observational Study in the THIN Database

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08 November 2008

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I. Specific Aims

Primary Aim 1: test the hypothesis that in patients with heart failure (HF), digoxin use is associated with higher mortality in women but not in men (i.e., that gender is an effect modifier for the association of digoxin use with mortality).

Primary Aim 2: test the hypothesis that effect modification between digoxin use and gender on mortality hazard is attenuated at lower dosage levels (i.e., that there is a three-way effect modification between gender, digoxin, and dose).

Secondary Aim 1: test the hypothesis that digoxin serum levels mediate the gender-digoxin interaction affecting mortality (in the subset of patients for whom these data are available)

II. Background

Several interventions in cardiovascular medicine may have different safety/efficacy profiles depending on gender. For example, digoxin may increase mortality by approximately 20% in women but not in men (1).

The initial signal of this differential effect by gender was identified as part of a post-hoc analysis of the Digitalis Investigation Group (DIG) trial (1). In absolute terms, women assigned to digoxin had a 4.2% higher risk of mortality compared to placebo while men had assigned to digoxin had a 1.6% lower risk of mortality compared to placebo. After multivariable adjustment, the relative risk of mortality in women was statistically significantly increased by digoxin use and the test for an interaction between sex and digoxin affecting mortality was significant at P = 0.014, an effect most likely mediated by higher serum levels in women (2).

The study was accompanied by an editorial which raised three complications in interpreting these results(3). The first was that the finding was post-hoc and could be a spurious false positive. The second letter was that there were significant differences at baseline between men and women (women had higher rates of cardiomegaly, severe heart failure, diabetes, and idiopathic heart failure; men had higher rates of heart failure related to myocardial ischemia and infarction) which could lead to different mortality rates by gender. The third was that, since one of the primary results of DIG had been that digoxin might be associated with increased mortality when serum concentration was greater than 1.0 ng/ml and with reduced mortality when serum concentration was lower than 1.0 ng/ml, it was reasonable to hypothesize that higher serum concentrations in women might be driving the differential effect on mortality by gender. Subsequent letters to the editor of the New England Journal of Medicine elaborated on these points (4, 5, 6). One letter looking at hospital registry data on Sweden found that in clinical practice digoxin serum levels did tend to be higher in female patients and were more often above the therapeutic range, lending credibility to the hypothesis that, if real, the differential mortality might be due to differences in serum level (7).

A different group of investigators subsequently carried out another post hoc analysis of the DIG trial, which examined mortality in women in the control arm versus women in the digoxin arm and stratified by serum digoxin concentration (2). This study found that the hazard ratio for death favored digoxin users (0.8, 95% CI 0.62-1.13) in women with a serum digoxin concentration up to 0.9 ng/ml, while women with a serum concentration of 1.2 ng/ml or greater

had a hazard ratio of 1.33 (95% CI 1.001-1.76) for death. This study further supported the hypothesis that serum digoxin concentration differences between the genders might be driving the sex-digoxin interaction. However, it was not powered to exclude the possibility that gender interaction might be present even after digoxin serum concentration was accounted for.

Finally, an observational study was conducted on patients enrolled in the Studies of Left Ventricular Dysfunction (SOLVD) trial to test for a sex-digoxin interaction affecting mortality, and found no evidence for such an interaction (7). The reason for this disagreement with the DIG post-hoc analysis (1) was not clear, although there were several possible explanations including the fact that the DIG study randomized digoxin use while the SOLVD study did not. Variables including digoxin dose and digoxin serum levels, which might have shed light on the discrepancy, were not included in the analysis. The SOLVD paper ended with the recommendation that further observational studies in other populations be conducted (7).

Further investigation of the potential sex-digoxin (or sex-digoxin-serum-level) interaction is clinically important because digoxin is still widely used by both women and men (8). It remains unclear whether digoxin should be used differently in the different genders (7) and recent publications warn against the use of digoxin in women (9)—an attitude which may unnecessarily curb use of a drug which has been shown to reduce hospitalizations in heart failure. It is plausible that careful maintenance of low digoxin serum concentration would prevent excess mortality in either gender (3), but it is not known whether current clinical practice avoids causing excess mortality in women.

We propose a longitudinal observational study in the THIN database to help answer this question, with prespecified outcomes looking both at gender-digoxin interaction on mortality and at gender-digoxin-dose interaction on mortality. The key advantages which THIN can offer over previously published research include a much larger sample size, use of data from routine clinical practice rather than the artificial environment of a clinical trial, the ability to adjust for digoxin dose within that large sample, and the ability to observe trends over a very long period of use (from the early 1990s up until 2007). Availability of digoxin serum levels on a minority of patients will add further value to the study. If there is a differential effect on mortality by gender, exploration of dose-dependency and serum concentration dependency will give insight into whether there is a way to avoid such effects. Indeed, as the average dose of digoxin has declined over time (8), a longitudinal database may allow a natural experiment to assess whether there was a differential effect by gender in the past which no longer exists under modern dosing practices. Since concerns about digoxin use in women continue to appear in the literature (9), either a positive or a negative finding would help guide clinical decision making, as long as confounding and other forms of bias are acceptably controlled.

III. Research and Methods

A. Overview

Using a retrospective cohort design, we will assess whether the mortality risk for digoxin users interacts with gender. We will also assess whether any interaction depends on the digoxin dose or has changed over the years. The database to be used (THIN, see below) provides

laboratory values on some patients, so it should also be possible to do a subgroup analysis adjusting for serum concentration of digoxin.

The clinical benefit of this study is that it will identify whether digoxin, as it is really being used, is harming women. Exploration of dose-dependency and serum concentration dependency will also give insight into whether there is a way to avoid such effects. Since concerns about digoxin use in women continue to appear in the literature, either a positive or a negative finding will help guide clinical decision making.

B. Rationale for Study Design

The proposed study design is a retrospective cohort study to estimate the hazard-ratio of all-cause deaths in patients chronically taking digoxin versus patients not taking digoxin. A retrospective observational cohort study was chosen for two reasons. First, while a randomized trial provided the initial evidence that digoxin may pose different mortality risks by gender, no randomized set of patients is available with sufficient numbers of outcomes to address all the aims of this protocol. Second, we wanted to study the effects of digoxin on mortality as it has actually been used in ordinary patients over time. This made data collected from routine clinical practice more relevant than data from randomized controlled trial data, provided problems with confounding and bias can be adequately addressed.

Confounding will be an issue in the design of this study, but the problem of confounding will be attenuated in two ways. The most problematic comparison would be to assess the direct effect of digoxin on mortality, since it is quite likely that patients who receive digoxin differ systematically from those who do not. However, the first primary outcome of this study is an assessment of drug-gender interaction. For this comparison to be confounded, a baseline difference between digoxin users and non-users would have to also differ by gender, a less likely scenario. Furthermore, the second primary outcome of this study is an assessment of drug-gender-dose interaction. Changes in dosing practices for digoxin are unlikely to be influenced strongly by gender; indeed, there is a strong secular trend towards lower doses of digoxin (8), which suggests that much variation in dosing practices is driven by factors other than patient characteristics.

C. Source Population/Database Description

The research design for this study is a retrospective cohort study. The source of the cohort will be a database from the United Kingdom called THIN (The Health Improvement Network). This database contains over 5 million individuals and has been validated for a variety of outcomes (10). The analysis will be restricted to individuals with HF, with digoxin as the exposure of interest, and assess hazard ratios for several relevant outcomes (all cause mortality as the primary outcome, with sudden cardiac death, digoxin toxicity, and hospitalization as secondary outcomes). THIN provides lab values on a subset of patients, so it should also be possible to execute a subgroup analysis adjusting for actual serum concentration of digoxin.

At the Center for Clinical Epidemiology and Biostatistics (CCEB), THIN can be used at minimal cost. A random 10% sample of the whole database is available for pilot work and can be used immediately for refinement of power calculations and development of analytic code for this

project. The primary author is familiar with the software packages (particularly MySQL and SAS) needed for this and will write the analytic code. The SAS package in particular includes procedures for the COX proportional hazard regression that will be used in the primary analysis (see next section).

D. Subjects

The analysis will be restricted to individuals with heart failure (HF). As detailed below, subjects must be treated with medication for this chronic disease.

1. Exposed Population

Digoxin exposure will be defined as consecutive, recurrent prescriptions (2 or more) of digoxin. Individuals will contribute exposed person-time from 90 days after their first prescription of digoxin until 30 days after their last prescription.

Individuals will contribute exposed person-time from the date of their first prescription of digoxin until 30 days after their last prescription. Exposure for this study is meant to capture current use of digoxin.

A last prescription is defined as a prescription not followed by another within 90 days. To clarify, if somebody takes digoxin for many years but during that time has a single gap of greater than 90 days, they will contribute two blocks of exposed time.

Of note, individuals in the control population are required to be on other medications associated with HF in order to contribute followup time. A parallel requirement is not in place for the cases (who contribute person-time even if digoxin is their only medication) in the primary analysis. There is a possibility that this would create bias by allowing more individuals with less severe or symptomatic HF into the case arm. However, in a secondary analysis, cases will be restricted to those with another medication in addition to digoxin.

At least one diagnostic code for heart failure will be required for any case.

2. Control Population

The control population will consist of individuals with HF who are being treated with medication for that condition. Only person time during such medical treatment (or for 30 days after the last prescription) will be included in the study. The reason for this rule is that a diagnostic code for HF unaccompanied by any medical treatment is likely to reflect either a mistaken code; an extremely mild case, or a patient so ill that they cannot adhere to chronic outpatient medication. Chronic medication use also implies frequent followup with a physician such that morbidities and mortality will be appropriately recorded. Medications commonly used chronically for heart failure include beta-blockers, diuretics (including aldosterone antagonists for severe cases), and ACE inhibitors.

Because all of these medications have other indications in addition to heart failure, at least one diagnostic code for heart failure will be required for any control. The diagnostic code must appear within 6 months of initiation of medication, to minimize ambiguous situations in which use of a medication like a beta-blocker may precede the onset of heart failure.

3. Exclusion Criteria

The only exclusion criterion for this study is that patients must have one year or more of follow-up time in the database prior to their incident diagnosis.

E. Covariates

Covariates in the analysis will include calendar year, age, sex, BMI and smoking status, (39% missing data, so BMI and smoking will only be included in a sensitivity analysis), history of hypertension, history of MI, history of stroke, history of hospitalization, and history of diabetes. Baseline statin use will also be used as a covariate. The most useful covariates (ie, consistently available and associated with the outcome) are likely to be medication use. The cornerstones of HF pharmacotherapy are beta-blockers, ACE inhibitors, and diuretics. These drugs will be entered as covariates and, numbers permitting, cases and controls will be matched by medication regimen. Diagnostic codes defining ejection fraction, severity of HF, or type of HF (systolic versus diastolic) are unlikely to be available on enough patients to be used in any but secondary analyses. Please see code appendix for additional details.

Baseline covariates will be assessed in the year prior to the index date for the study. For both cases and controls, the index date is the date of initiation of medical treatment for HF. Medication exposure at baseline consist of the constellation of medications intiated before or within 90 days of the beginning of treatment for HF. Medication exposure will also be analysed as a time-varying covariate as a sensitivity analysis; if this sensitivity analysis alters the study conclusions then more sophisticated methods for analyzing time varying covariates, such as a marginal structural model, will be employed.

F. Outcome

The primary outcome for this study is death, which is recorded in the THIN database as a death date. Secondary outcomes, such as death from cardiac arrhythmia and digoxin toxicity, will be defined using medical diagnosis codes (see code appendix).

G. Codes

Medical and drug codes were identified through text searches of the THIN medical codes databases, hand screening, and consultation with other researchers at the University of Pennsylvania who have done research on digoxin and on HF. The codes are included as an appendix.

H. Analysis

The initial analysis will be descriptive, comparing exposed and unexposed individuals. Differences between groups will be tested for significance using chi-square and t-tests depending on whether the variable of interest is dichotomous or continuous. Incidence rates in each group will be calculated, then hazard ratios will be calculated using Cox proportional hazards models. Both unadjusted and adjusted models will be run. Adjusted models will include all variables outlined in the Covariate section above.

The final analysis to address the first primary aim will most likely be presented as two multivariable Cox models, one for males and one for females. The formal significance test for a digoxin-gender interaction, however, will most easily be done via a single Cox model which includes sex, digoxin exposure, and a sex-digoxin exposure interaction term in the model.

To address the second primary aim, average daily dose of digoxin will be calculated and the analysis will be stratified by quartiles of average daily dose. Because of the strong secular downward trend in digoxin dose with time, it will be particularly important to control for calendar year in this analysis. In an exploratory component of this aim, we will investigate whether calendar year, possibly in combination with physician prescribing preference, can be used as an instrumental variable and whether this adjustment changes the results of the analysis. In theory, an instrumental variable can control for confounding that is not necessarily captured by conventional multivariable adjustment (11).

To address the third (and secondary) aim, we will identify patients for whom digoxin serum levels are available in the database. For that subset of patients, the analyses described for aim 1 can be repeated but including serum level of digoxin in the model. It is possible that there will be insufficient data of this kind for us to complete this aim.

In all aims, sensitivity analysis for the potential effect of unmeasured confounding will also be conducted (12).

I. Sample Size

Pilot data indicate that approximately 800,000 patients in the THIN database have had multiple prescriptions for digoxin at some point and that 30,000 of these patients have died. While these pilot numbers do not incorporate exclusion criteria or assess how many of these deaths occurred while the patient was taking digoxin, they suggest that the number of eligible outcomes is likely to be large and will certainly support aims 1 and 2. Even if only 1% of these deaths are eligible outcomes in digoxin-exposed individuals, the study should be 90% powered at an alpha of 0.05 to detect a hazard ratio for the digoxin-gender interaction of 1.3 (analysis done using Schoenfeld's equations for time-to-event analysis).

J. Plan for reporting and following up results

This project is intended for peer-reviewed publication. The primary parameter estimates from this research will be a hazard ratio for a digoxin-gender interaction (primary aim 1, regarding the basic question of whether such an interaction is present). The results of primary aim 2 will be more complicated to present but we anticipate reporting multiple digoxin-gender interaction hazard ratios after stratification by calendar year.

K. Anticipated Limitations

●Representativeness of Cohort: Since exposed and unexposed are drawn from a large population sample and treated as a cohort, they should be representative of the UK population.

●Misclassification of Exposure: The exposure is use of a prescription drug. Prescriptions issued are automatically captured by the systems that contribute data to THIN and these data are considered highly reliable. As a proxy for use of the drug, this is not a perfectly valid measure because patients may never fill or take the prescription. However, such misclassification should be minimal and nondifferential. Dosing levels will be computed from prescription data as has been done in other studies (8); similar concerns about nondifferential misclassification apply.

●Misclassification of Outcome:

○Mortality is recorded in THIN as a mortality code. The specificity of this measure should be good and can be confirmed by checking to make sure that medical followup ends at the time of reported death. Sensitivity may be imperfect if some deaths are not recorded and merely appear as the end of followup in THIN; however, this is unlikely to be a major problem and would most likely result in nondifferential bias to the null.

●Confounding by Indication: confounding by indication is likely to be a less severe problem because this study focuses on interactions rather than on the main drug effect. Adjustment for covariates will also be employed.

IV. Implications

This project has implications for basic pharmacologic science, clinical practice, epidemiological methods, and future research.

A. Basic Science

The major basic science contribution of this project would be to assess whether there is a dose-gender-digoxin three-way interaction. A dose-response relationship, which is unlikely to be due to confounding, would support the current hypothesis that differential mortality is due to the same dosing level resulting in different serum concentration in the different genders.

B. Clinical Practice

This project is focused on clinical practice: the question is whether there is a need for different usage guidelines for digoxin based on gender. In particular, primary aim 2 will address the hypothesis that modern dosing practices equalize the risk between genders.

C. Epidemiological Methods

This project will result in careful examination of two methodological questions. The first is whether interactions are typically less confounded than direct effects. The second is whether a strong secular trend in how a drug is used can lead to a convincing demonstration of how the changing practice impacts outcomes.

V. Timeline and Logistics

A. Human Subjects Protections

1. Approval from Penn IRB
2. Approval from ethics bodies governing THIN

B. Grant Support

For support for this study, I will apply to the CTSA-ACARD Internal Small Grant Program (attached). It is possible to apply for any of 5 levels of support in working with the THIN database. For this project, the most appropriate is level 4.

Level 4 support includes exporting data files from THIN to a PC environment. ACARD intends to fund small grants to export analytic data files from the full THIN (residing on a server) for use on a PC. This funding is for researchers who have pilot-tested their variable creation, cohort selection criteria, and statistical code using the 10% sample data residing on a dedicated PC (known as 'Victoria'). These grants are for $1000 each, all of which must be used for the cost of the Biostatistical Analysis Center (BAC) to export the data from the full THIN database for further analysis by the investigators. The BAC will implement the SQL, SAS, and/or STATA code provided by the applicant to create the study data files. The BAC will not be responsible for determining the integrity of the code. If selected for funding, the funds will be directly transferred from ACARD to BAC.

I am writing my own SQL, SAS, and STATA code to conduct this analysis with the guidance of my thesis advisor and my biostatistics advisor. PC resources available within the CCEB will be sufficient to implement this code.

C. Timeline

To do	*Sep. '08*	*Oct.*	*Nov. '08*	*Dec.*	*Jan.*	*Feb.*
Develop protocol	**********					
Penn IRB approval (done)		********				
UK Ethics Approval			**********			
Write, pilot, and debug SAS/SQL code			**********	**********	**********	
Data extraction and						*

analysis	
Writeup primary results	*****

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Figure 1a. Trends in Drug Use Among Men

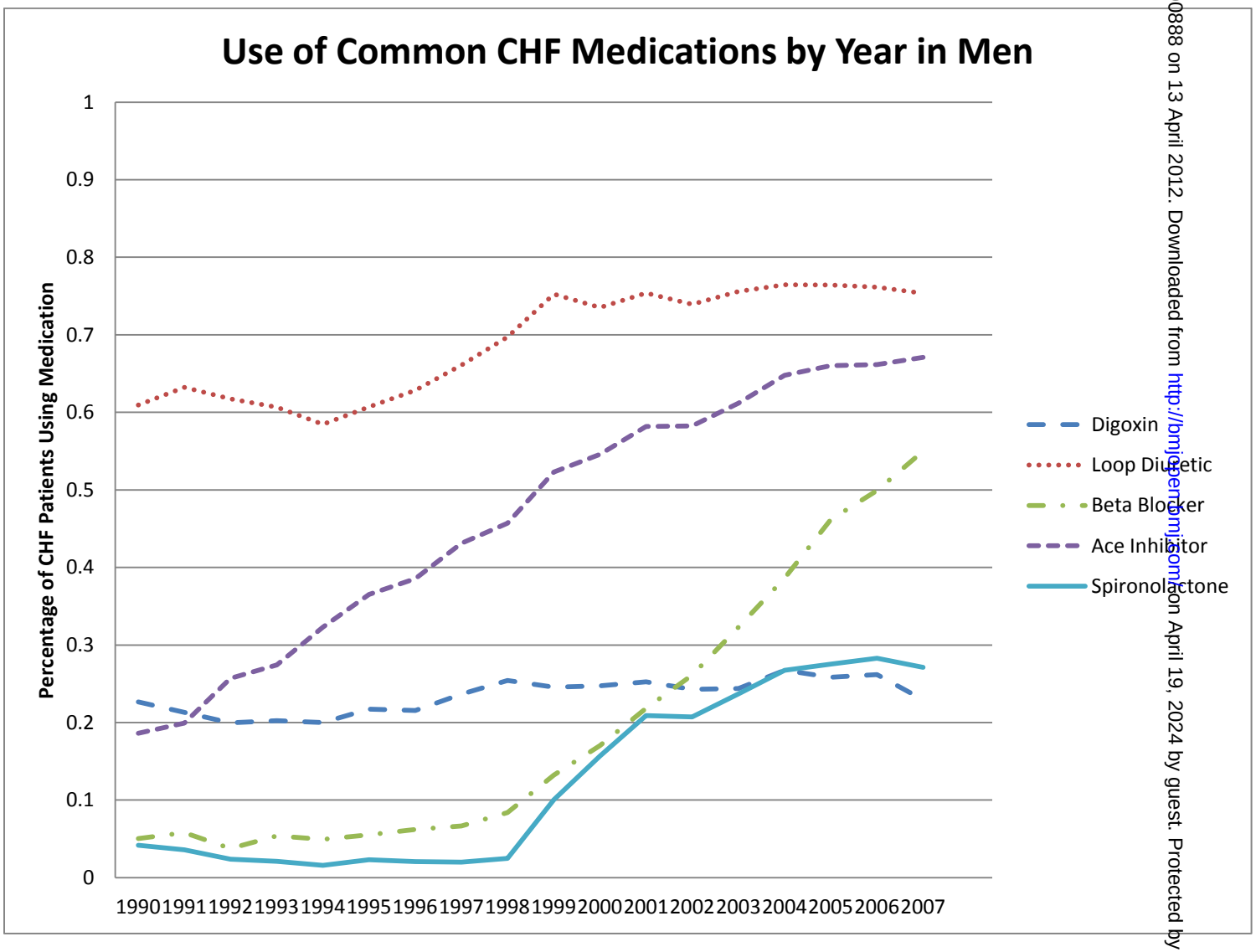
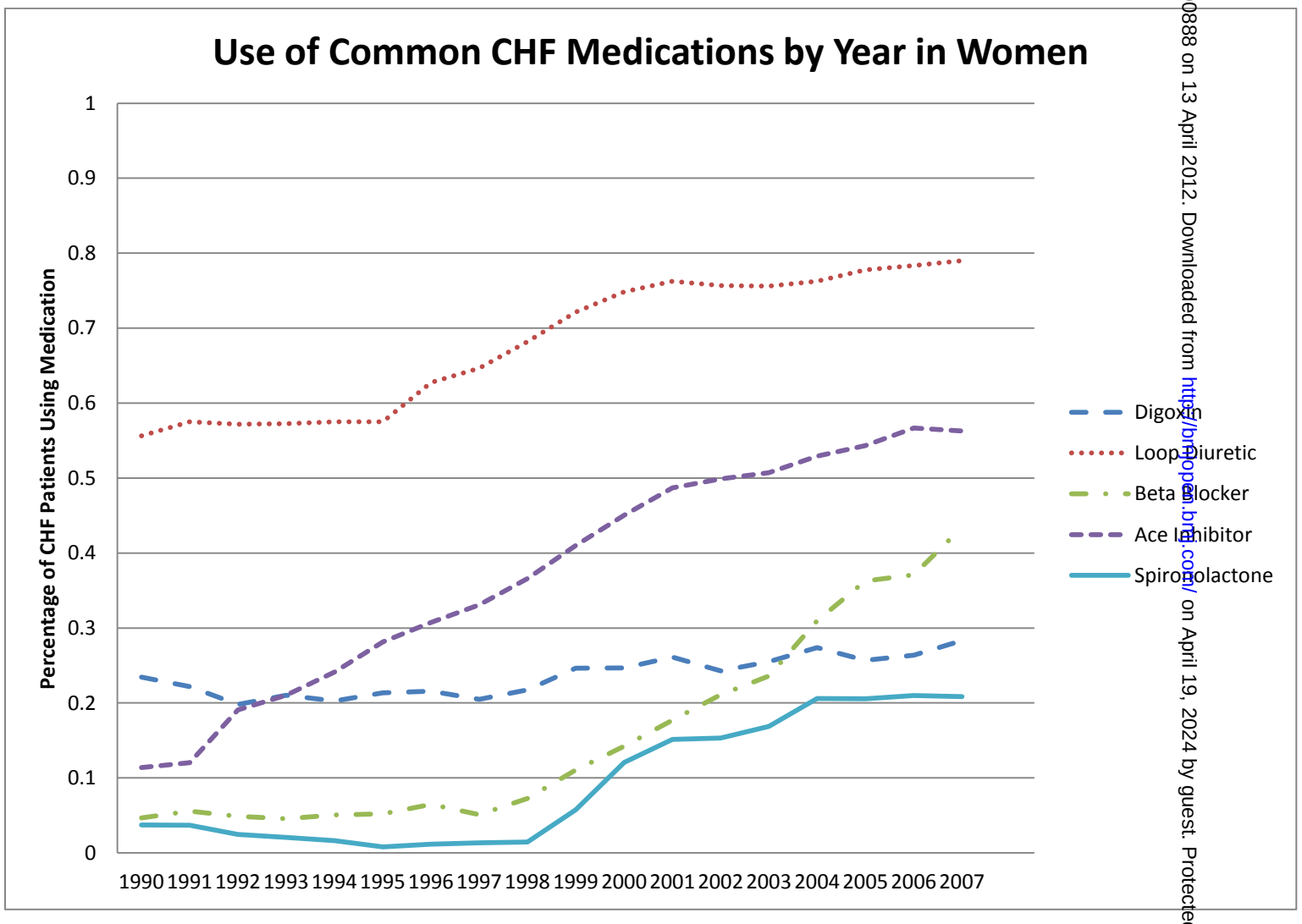


Figure 1b. Trends in Drug Use Among Women



Correction

Flory JH, Ky B, Haynes K, *et al*. Observational cohort study of the safety of digoxin use in women with heart failure. *BMJ Open* 2012;**2**: e000888. The fourth author in this article should be listed as Brunelli SM (not S Brunelli M).

BMJ Open 2012;**00**:e000888corr1. doi:10.1136/bmjopen-2012-000888corr1