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Digitoxin medication and cancer; case control and internal dose-response studies

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Abstract

Background: Digitoxin induces apoptosis in different human malignant cell lines in vitro. In this paper we investigated if patients taking digitoxin for cardiac disease have a different cancer incidence compared to the general population.

Methods: Computer stored data on digitoxin concentrations in plasma from 9271 patients with cardiac disease were used to define a user population. Age and sex matched controls from the Norwegian Cancer Registry were used to calculate the number of expected cancer cases.

Results: The population on digitoxin showed a higher incidence of cancer compared to the control population. However, an additional analysis showed that the population on digitoxin had a general increased risk of cancer already, before the start on digitoxin. Leukemia/lymphoma were the cancer types which stood out with the highest risk in the digitoxin population before starting on digitoxin. This indicates that yet unknown risk factors exist for cardiovascular disease and lymphoproliferative cancer.

An internal dose-response analysis revealed a relationship between high plasma concentration of digitoxin and a lower risk for leukemia/lymphoma and for cancer of the kidney/urinary tract.

Conclusion: Morbidity and mortality are high in the population on digitoxin, due to high age and cardiac disease.These factors disturb efforts to isolate an eventual anticancer effect of digitoxin in this setting. Still, the results may indicate an anticancer effect of digitoxin for leukemia/lymphoma and kidney/urinary tract cancers. Prospective clinical cancer trials have to be done to find out if digitoxin and other cardiac glycosides are useful as anticancer agents.

Background

Cardiac glycosides have been used in the treatment of cardiac disease for more than 200 years. In most Western countries digitoxin has been replaced by digoxin and other drugs. Digitoxin is still today the most common cardiac glycoside prescribed in Norway [1]. Digoxin has a shorter elimination half life and is often regarded easier to dosage than digitoxin. However, more attention is again paid to digitoxin as a valuable cardiac drug, espe-

cially for the eldery, and perhaps its use will increase in the future [2].

Cardiac glycosides also have well known antiproliferative effects on tumor cells [3–5]. Some cardiac glycosides have been evaluated in short term animal models. The conclusion from these experiments is that very high doses, probably toxic, would be needed for obtaining anticancer effects in humans [6]. In contrast, we have previously found that non toxic concentrations of digitoxin and digoxin inhibits growth and induce apoptosis in different human malignant cell lines, whereas highly proliferating normal cells were not affected [7–9]. The capability of cardiac glycosides to induce apoptosis has recently been confirmed in other studies [10,11]. There is a great difference in susceptibility for cardiac glycosides in different species indicating that one can not extrapolate the results from animal models into humans [4].

In our in vitro experiments the apoptosis-inducing effect was more potent for digitoxin than for digoxin, and for digitoxin there was a dose response pattern; the higher concentration the more apoptosis. Another recent report on the anticancer effects of different cardiac glycosides on tumor cell lines also confirms that digitoxin seems more potent than digoxin [12].

As previous studies on cancer risk in patients on digitalis more or less exclusively concern digoxin [13–16] we have studied the possible anticancer effect of digitoxin in patients with cardiac disease. Thus, we wanted to examine if the strong anticancer effects detected in vitro were evident in vivo in a patient population on the drug for cardiac disease.

Material and Methods

In Norway patients on digitoxin usually have their plasma concentration checked shortly after the initiation of the treatment. The basis of the study is all cardiac patients (n = 9271, 5026 women and 4245 men) who had their first digitoxin concentration measurement carried out in the period 1986–96 at the University Hospital of Trondheim. The settlement is very stable for these patients, so our figures are not influenced by that patients have had their plasma digitoxin measured in any other hospital. The digitoxin concentrations were measured by a radio-immunoassay method (Coat-A-Count Digitoxin, Diagnostic Products Corporation, Los Angeles, USA). The reference range for therapeutic plasma digitoxin concentration at our laboratory is 15–33 nmol/1 (12–25 ng/ml). The mean age for the total digitoxin population was 75.8 years (1 SD = 10.2), for men 73.3 years (1 SD = 10.3) and women 78.0 years (1 SD = 9.5). After approval of the Norwegian Data Inspectorate, the regional ethical committee and the Norwegian Health Inspectorate, the digitoxin data were linked to individual data on cancer in the population based Norwegian Cancer Registry.

To study the association between digitoxin use and cancer, three different approaches were used.

Firstly, a prospective design was chosen. All the digitoxin users with no prior cancer diagnosis formed a basis cohort from the time they have their first digitoxin plasma concentration measurement carried out. This cohort was followed for the occurrence of cancer until the subjects died or up to December 31, 1996. The expected number of cases for each type of cancer was calculated by applying the national cancer rates matched exactly on year of birth, age and sex. The standardized incidence ration (SIR) was calculated for all types of cancers having more than 30 expected cases. The 95% confidence intervals were estimated on the basis of the Poisson-distribution.

The second approach was a retrospective design based on persons who were identified in the laboratory database as digitoxin users. For each becoming digitoxin user one control person was randomly picked out from the general population. These controls were matched by birth date and gender and they should be alive at the time when the corresponding becoming digitoxin user started on digitoxin.These two groups were compared with respect to having a cancer diagnosis during the period of observation and the Odds ratio of cancer cases before start on digitoxin was used as a risk estimate. The 95 % confidence intervals were estimated on the basis of the Mantel-Haenzel chi-square estimation. As the control persons are randomly picked out from the general population matched by birthday and gender, we do not know anything about eventual drug use in the controls. Thus, some of the controls might use digitoxin and other drugs. However, the frequency of digitalis users in the general population is so low that it is not likely to corrupt our figures significantly.

The third approach was a prospective internal dose-response on the cohort on digitoxin users where the risk of cancer was studied by different levels of digitoxin plasma concentration at first measurement divided in tertiles and a Cox regression analysis was carried out. For the trend analysis the continuous values for the digitoxin variables was used.The the lowest plasma digitoxin concentration (< 16 ng/ml) was set as reference.

Results

Table 1 demonstrates a general higher cancer incidence in the population on digitoxin compared to what is expected in the general population. However, it is not clear whether the population of digitoxin users differ from the general population in cancer risk already before start on

Table 1: Observed and expected cancer cases among digitoxin users.

ICD-10	Site	Sex	Observed	Expected	SIR (95 % CI)
C50	Breast	F	57	45.7	1.25(0.95, 1.62)
C61	Prostate	M	108	86.7	1.25(1.03, 1.50)
C18–21	Colo-rectal	M+F	127	98.5	1.29(1.06, 1.51)
C32–34	Lung	M+F	63	46.4	1.35(1.04, 1.74)
C64,C65,C67,C68	Kidney, urinary	M+F	59	51.8	1.14(0.87, 1.47)
C43.C44	Melanoma, other skin	M+F	61	49.7	1.23(0.94, 1.58)
C81–C85,C88–C92	Leukemia/Lymphoma	M+F	53	37.5	1.41 (1.06, 1.85)
C00–C97	All sites	M+F	641	502.8	1.27(1.18, 1.37)

Observed and expected cancer cases amoung digitoxin users, and standardized incidence ratio (SIR) presented for all types of cancers having more than 30 expected cases. The cancer types are divided in the groups according to ICD-10 (International Classification of Disease).Observed=cancer cases in the population on digitoxin after start on the drug,no lag time applied. Expected=expected cancer cases calculated by applying the national cancer rates matched exactly on year of birth,age and sex.

Table 2: Number of cancer cases among subjects that subsequently became digitoxin users.

ICD-10	Site	Sex	Digitoxin group	Control group	Odds Ratio (95% CI)
C50	Breast	F	275	232	1.19(1.11, 1.28)
C61	Prostate	M	370	296	1.25(1.08, 1.45)
C18–21	Colo-rectal	M+F	430	325	1.32(1.15, 1.52)
C32–34	Lung	M+F	163	112	1.46(1.15, 1.86)
C64,C65,C67,C68	Kidney, urinary	M+F	201	180	1.06(0.95, 1.17)
C43.C44	Melanoma, other skin	M+F	154	191	0.81 (0.66, 0.99)
C81–C85,C88–C92	Leukemia/Lymphoma	M+F	198	117	1.69(1.35,2.11)
C00–C97	All sites	M+F	2435	2017	1.21 (1.15, 1.28)

Number of cancer cases among subjects that subsequently became digitoxin users,and among matched controls.Digitoxin group=the cancer cases among the 9271 patients constituting the database just before the start on digitoxin medication. Control group=for each subject in the digitoxin group one control was randomly picked out from the general population. These controls were matched on birthday and gender and should be alive at the time when the corresponding digitoxin user started on digitoxin.

the drug. Table 2 presents the results of the second approach where we wanted to examine if the persons on digitoxin had an increased risk of cancer already before they had started to take the drug. Here the actual number of cancer cases in the population of "becoming digitoxin users" is compared with the number of cases in the age and sex matched control group (presented as Odds ratio (OR) for each type of cancer). Thus, more cancer cases were seen in the group to become digitoxin users (OR > 1.00) except for melanoma and other skin cancers. Lymphoma and leukemia are presented as a single group in the tables due to the close relationship between them. In the prospective part of the study (as presented in Table 1) for leukemia alone SIR was 1.39 (95% CI: 0.95–1.97) (31 observed versus 22.3 expected). In the comparison between becoming digitoxin users and the controls (as presented in Table 2), the Odds ratio for leukemia was 1.95 (95% CI: 1.42–2.67). The internal dose-response analysis is shown in Table 3. For most types of cancer we could not find any association, neither for all types of cancer combined. However, a test for trend by level of digitoxin concentration indicated a trend toward a protective effects of high digitoxin levels for the lymphoma/leukemia group ($p = 0.008$) and for kidney/urinary organ cancers ($p = 0.05$).

Discussion

The intention with the present study was to investigate whether use of digitoxin induces any changes in the incidence of cancer. One strength in our material is that we

use plasma concentration measurements for defining the digitoxin using population. In previous studies drug use has been estimated by employing dispensing data or questionnaires about drug use [13–15]. These methods are less reliable than measuring plasma drug concentration, as neither completely account for variations in recall biases, complicance, nor for the existing inter-individual pharmacokinetic variability.

As mentioned earlier, digoxin and other cardiac glycosides are more commonly used in many countries and the use in the earlier studies is usually not specified to just one drug, but to the digitalis group of prescription drugs [13–15,17]. We found a general higher incidence of cancer among the patients on digitoxin. This is in agreement with the earlier published studies [13–15,17]. These studies have just presented figures on cancer incidence after commencement on the drug, as we have done in the prospective part of our study (Table 1). The general higher incidence of cancer in the cardiac population on digitoxin thus prompted us to perform the analysis presented in Table 2. These figures show that the population to become digitoxin using has an increased risk of having cancer also before starting on digitoxin medication. Thus there exist risk factors associated with both cancer and cardiovascular disease. For some types of cancer this association may be explained by food habits, smoking and other factors. Intake of food rich in antioxidants and a favorable composition of carbohydrates, fats and proteins as well as vitamines and minerals may reduce morbidity in both cardiovascular disease and cancer [18]. Smoking is an evident risk factor for both lung cancer and cardiovascular disease.

Surprisingly, the leukemia/lymphoma group shows the highest Odds ratio and melanoma/other skin cancers the lowest (Table 2). Excessive exposition to sunlight shows a positive correlation with the development of skin cancer [19]. The lower Odds ratio (OR) for the development of skin cancer could hypothetically be explained by a different life style, as people who will develop cardiac disease and subsequent become digitoxin users may be more in house and therefore not exposed to sunlight to the same extent. However, the increased SIR value for skin cancer (Table 1) is more difficult to explain.

The high risk of lymphoproliferative cancers before start on digitoxin is intriguing (Table 2). Microbiological agents are associated with some types of cardiovascular disease and also for the development of cancer, but whether mutual agents exists are unclear [20,21]. Our ratios are well in agreement with a previous study focusing on drug use and other factors preceding non Hodgkin lymphoma [15]. This substantiates that yet unknown common risk factors exist for the development of cardiovascular disease and lymphoproliferative cancer. A possible protective effect of digitoxin here is indicated by a dose response pattern in the internal analysis. In addition, human leukemic cell lines in the form of Jurkat (T-cell leukemia) and Daudi (B-cell leukemia) are among the most susceptible for apoptosis induction by digitoxin treatment in vitro [8]. This further support that the SIR – OR ratio (1.39/1.95) for leukemia of 0.71 may indicate an anticancer effect.

It is evident that tracking anticancer effects in a population of patients with serious cardiac disease is hampered by several confounding factors. The mean time of digitoxin use before the cancer diagnosis is just about 3 years in our material. In general studies on cancer incidence in relation to drug use may be biased by that the cancer, before detected, induces symptoms treated by the drug of study. Moreover, as patients seek medical advice for cardiac problems they are also subjected to a more careful physical examination and a tumor may therefore be detected and diagnosed soon after the start of treatment for cardiac disease, thus leading to the false impression that the drug induced the cancer. This can be avoided by introducing lag times in the analysis. However in view of the mostly unknown time course for the occurrence and initial progression of solid tumors before they become detected, the lag time applied will be arbitrary chosen. One year lag time did not have any impact on our figures indicating that this effect has not corrupted the presented results. Another potential confounding factor is the use of other cardiac drugs in the digitoxin group. Most likely, the use of drugs such as angiotensin converting enzyme (ACE) inhibitors, calcium antagonists and warfarin is higher in the digitoxin group than in the control group. The use of ACE inhibitors and warfarin have been associated with a decreased risk of cancer [22,23], whereas the use of calcium antagonists has been associated with an increased risk of cancer in some studies, but not in all [24]. Thus, use of these drugs might have affected the results in both directions. Unfortunately, information on concomitant drug use was not available in the database.

Chemotherapy, especially in the form of anthracyclines, may induce cardiac congestion and thus subsequent use of digitalis [25]. However, it seems that it is just a small fraction of all patients on digitalis who has had the cardiac disease as a result of chemotherapy, so this should not have corrupted our figures (Table 2), but still it will counteract the detection of an eventual anticancer effect of digitoxin to some extent in the analysis.

Based on theoretical assumptions it may be that the cancer incidence is not dramatically changed in patients on digitalis, but that the survival nevertheless could be af-

Table 3: An internal dose-response analysis.

ICD-10	Site	Digitoxin concentration ng/ml		
		<16	16–22	>22
C50	Breast	1.00 (reference)	1.04(0.59, 1.84)	0.90(0.48, 1.67)
C61	Prostate	1.00 (reference)	0.79(0.51, 1.22)	0.89(0.56, 1.40)
C18–C21	Colo-rectal	1.00 (reference)	0.97(0.66, 1.42)	0.92(0.61, 1.40)
C32–C34	Lung	1.00 (reference)	1.25(0.71,2.22)	1.06(0.57,2.00)
C64, C65 C67, C68	Kidney, urinary organs	1.00 (reference)	0.30(0.15,0.57)	0.45 (0.24, 0.83)
C43,C44	Melanoma, other skin	1.00 (reference)	1.16(0.60,2.25)	1.29(0.65,2.57)
C81–C85 C88–C92	Leukemia and Lymphoma	1.00 (reference)	0.67(0.38, 1.18)	0.57(0.30, 1.08)
C00-C97	All sites	1.00 (reference)	0.84(0.71,0.99)	0.86(0.72, 1.02)

An internal dose-response analysis. Age-adjusted relative risks (95% confidence limits) of specific types of cancer by concentration levels (tertiles) of digitoxin. A Cox regression analysis was used.

fected. Our material was not suited for survival analyses due the high age of the patients and the correspondingly high mortality due to cardiac disease. Cancer patients are also prone to get their cancer diagnosis on the death certificate, even if other conditions are the direct cause of death. To make a fair analysis with focus on survival time, autopsy data for most of the patients would be required [26], and such data were not available

Conclusion

It is evident from the present study that too many disturbing factors exist in a population with serious cardiac disease for making proper examination of possible anticancer effects of cardiac glycosides. However, our study gives some support to data from previous studies indicating a higher incidence of cancer detected in patients treated with digitalis, but this association should be ascribed to underlying factors inducing and/or promoting both cancer and cardiovascular disease and not the actual use of digitalis.

The possible role for cardiac glycosides in cancer treatment has to be evaluated in prospective clinical studies with cardiac glycosides as primary anticancer agents.

Competing interests

None declared

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References

1. Westheim A, Dickstein K, Gundersen T, Hole T, Kjekshus J, Myhre ES, Ronnevik PK, Samstad S, Smith P: **[Chronic heart failure – suggestion to a management program].** *Tidsskr Nor Laegeforen* 1999, **119**:3427-3431
2. Roever C, Ferrante J, Gonzalez EC, Pal N, Roetzheim RG: **Comparing the toxicity of digoxin and digitoxin in a geriatric population: should an old drug be rediscovered?** *South Med J* 2000, **93**:199-202
3. Shiratori O: **Growth inhibitory effect of cardiac glycosides and agly cones on neoplastic cells: in vitro and in vivo studies.** *Gann* 1967, **58**:521-528
4. Repke KRH: **The role of the Na+/K+ pump in normal and cancer cell proliferation.** *In: Biomemebranes, Basic and Medical Research (Edited by Benga G, Tager JM) Berlin, Springer Verlag* 1988161-176
5. Repke KR, Schon R, Megges R, Wetland J, Nissen E, Matthes E: **Potential suitability of Na+/K(+)-transporting ATPase in prescreens for anti-cancer agents.** *Anticancer Drug Des* 1995, **10**:177-87
6. Cassady JM, Suffness M M: **Terpenoid antitumor targets.** *In: Anti cancer agents based on natural product models (Edited by Cassady JM, Douros JD) New York, Academic Press* 1980201-269
7. Haux J: **Digitoxin is a potential anticancer agent for several types of cancer.** *Med Hypotheses* 1999, **53**:543-548
8. Haux J, Lam M, Marthinsen ABL, Strickert T, Lundgren S: **Digitoxin, in non toxic concentrations, induces apoptotic cell death in Jurkat T cells in vitro.** *Z Onkol* 1999, **31**:14-20
9. Haux J, Solheim O, Isaksen T, Angelsen A: **Digitoxin, in non-toxic concentrations, inhibits proliferation and induces cell death in prostate cancer cell lines.** *Z Onkol* 2000, **32**:11-16
10. Kawazoe N, Aiuchi T, Masuda Y, Nakajo S, Nakaya K: **Induction of apoptosis by bufalin in human tumor cells is associated with a change of intracellular concentration of Na+ ions.** *J Biochem (Tokyo)* 1999, **126**:278-286
11. McConkey DJ, Lin Y, Nutt LK, Ozel HZ, Newman RA: **Cardiac glycosides stimulate Ca2+ increases and apoptosis in androgen-independent, metastatic human prostate adenocarcinoma cells.** *Cancer Res* 2000, **60**:3807-3812
12. Johansson S, Lindholm P, Gullbo J, Larsson R, Bohlin L, Claeson P: **Cytotoxicity of digitoxin and related cardiac glycosides in human tumor cells.** *Anticancer Drugs* 2001, **12**:475-483
13. Friedman GD, Ury HK: **Initial screening for carcinogenicity of commonly used drugs.** *J Natl Cancer Inst* 1980, **65**:723-733

14. Friedman GD, Ury HK: **Screening for possible drug carcinogenicity: second report of findings.** *J Natl Cancer Inst* 1983, **71**:1165-1175
15. Bernstein L, Ross RK: **Prior medication use and health history as risk factors for non-Hodgkin's lymphoma: preliminary results from a case-control study in Los Angeles County.** *Cancer Res* 1992, **52**:5510-5515
16. Stenkvist B: **Is digitalis a therapy for breast carcinoma?** *Oncol Rep* 1999, **6**:493-496
17. Lenfant-Pejovic MH, Mlika-Cabanne N, Bouchardy C, Auquier A: **Risk factors for male breast cancer: a Franco-Swiss case-control study.** *Int J Cancer* 1990, **45**:661-665
18. McCarty MF: **Vegan proteins may reduce risk of cancer, obesity, and cardiovascular disease by promoting increased glucagon activity.** *Med Hypotheses* 1999, **53**:459-85
19. Armstrong BK, Kricker A, English DR: **Sun exposure and skin cancer.** *Australas J Dermatol* 1997, **38**:1-6
20. Cassell GH: **Infectious causes of chronic inflammatory disease and cancer.** *Emerg Infect Dis* 1998, **4**:475-87
21. Kinlen LJ: **High-contact paternal occupations, infection and childhood leukaemia: five studies of unusual population-mixing of adults.** *Br J Cancer* 1997, **76**:1539-45
22. Lever AF, Hole DJ, Gillis CR, McCallum IR, McInnes GT, MacKinnon PL, Meredith PA, Murray LS, Reid JL, Robertson JW: **Do inhibitors of angiotensin-I-converting enzyme protect against risk of cancer?** *Lancet* 1998, **352**:179-184
23. Schulman S, Lindmarker P: **Incidence of cancer after prophylaxis with warfarin against recurrent venous thromboembolism. Duration of Anticoagulation Trial.** *Engl J Med* 2000, **342**:1953-1958
24. Cohen HJ, Pieper CF, Hanlon JT, Wall WE, Burchett BM, Havlik RJ: **Calcium channel blockers and cancer.** *Am J Med* 2000, **108**:210-215
25. Brestescher C, Pautier P, Farge D: **[Chemotherapy and cardiotoxicity].** *Ann Cardiol Angeiol* 1995, **44**:443-447
26. Britton M: **Diagnostic errors discovered at autopsy.** *Acta Med Scand* 1974, **196**:203-210

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