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Taraxacum officinale extract induces antitumorigenic effects in ovarian carcinoma cell lines

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Summary

Purpose of Investigation: Patients suffering from ovarian cancer often use mistletoe preparations in combination with *Taraxacum officinale* (dandelion) extracts as supplementary therapy to conventional medicine. However, scientific data concerning antitumorigenic effects of *Taraxacum* extracts and co-treatment with mistletoe extracts are insufficient. *Materials and Methods:* Sensitivity of cancer cell lines towards a *Taraxacum* extract was determined by cell viability assays. Effects of *Taraxacum* on cell proliferation, cell cycle distribution, mitochondrial integrity, as well as migration and invasion were investigated in ovarian carcinoma cells (NIH:OVCAR-3 and SKOV-3). *Results: Taraxacum* extract caused a reduction of cell viability in a panel of adult cancer cell lines (cervical-, colon-, endometrial-, hepatocellular-, lung-, mamma-, ovarian-, pancreatic-, and urinary bladder carcinoma), whereby gynecological tumors were particularly susceptible. Inhibitory effects on cell viability, migration, mitochondrial integrity, and induction of apoptosis were observed in ovarian cancer cells. Combined therapy with mistletoe extracts revealed synergistic effects. *Conclusion:* This study provides first preclinical data on the antitumorigenic effects of *Taraxacum* and demonstrates the effectiveness of a combination treatment with mistletoe extracts in ovarian cancer cells.

Key words: Anthroposophic remedy; Gynecological tumors; Phytotherapy; Viscum album; Taraxacum officinale.

Introduction

Ovarian cancer is the most lethal of all gynecological cancers and the fifth most common deadly cancer disease (100,000 deaths per year) among women worldwide [1, 2]. A major clinical challenge of this disease is that most women are diagnosed late in the development of ovarian cancer, as early appearances of the disease causes minimal, vague or no symptoms at all [3]. Despite intensive multimodal treatment options including surgery, chemotherapies, radiation, hormone therapy, and targeted therapies, the five-year survival rate (46 %) unfortunately stagnates in the last decades [4].

In oncology treatment, complementary and alternative medicine (CAM) is often used supplementary to conventional therapies. Among ovarian cancer patients 44 % to 53 % receive CAM therapies and 23 % of these patients decide to try herbal therapies or other plant extracts [5].

In Europe, mistletoe (*Viscum album* L.*)* preparations are the most popular herbal remedies applied for tumor diseases. The beneficial effects of mistletoe extracts, such as improvement of life quality and prolonging of survival, are demonstrated for adult cancer diseases, including gynaecological tumors in several clinical studies [5-7]. It is supposed that lectins and viscotoxins are the major components responsible for the cytotoxic properties of mistletoe extracts [8, 9]. In CAM, particularly in hospitals with a focus on anthroposophic medicine, mistletoe extracts are combined with *Taraxacum officinale*, also known as dandelion [10]. *Taraxacum* is usually applied as a diuretic and as a remedy for gastrointestinal disorders [11]. Recent preclinical studies also demonstrated anti-tumoral effects of *Taraxacum officinale* (dandelion) on solid tumors such as prostate and breast cancer [12, 13], hepatocellular carcinoma [14], and melanoma [15].

In addition, inhibition of proliferation and induction of TP53 mediated apoptosis has also been shown in ovarian carcinoma cell lines upon treatment with ethanolic extracts of dried *Taraxacum officinale* leaves [16]. However, evidence for synergistic effects by combining mistletoe and dandelion extracts has not yet been established.

For the first time, the present authors investigated the anti-tumoral effects of an aqueous fermented *Taraxacum*

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extract on adult cancer cell lines with a focus on ovarian carcinoma cells. In addition, they intended to shed light on synergistic effects between *Taraxacum officinale* (dandelion) and *Viscum album* (mistletoe) extracts.

Materials and Methods

Taraxacum officinale extract, *Taraxacum e planta tota ferm 34c* (subsequently referred to as *Taraxacum*) was manufactured according to the official production method 34c, laid down in the official German Homeopathic Pharmacopoeia (GHP). In short, comminuted fresh flowering plant material undergoes a lactic acid fermentation process (7 d) followed by a maturation period of six months at 15°C. The aqueous extracts, *Iscucin Tiliae* and *Iscucin Pini*, are produced according to the GHP production method 38. Both extracts are derived from different host trees (lime tree and pine) and therefore differ in their spectra of ingredients. *Iscucin Tiliae* is characterized by high lectin levels.

Sensitivity to *Taraxacum* extracts was assessed in a cell line panel comprising 14 adult cancer cell lines from different entities and a non-tumorigenic human fibroblast cell line, NHDF-C. NIH:OVCAR-3, SKOV-3 (ovarian carcinoma), Ishikawa, AN3 Ca (endometrium carcinoma), EJ-28, RT-112 (urinary bladder carcinoma), A549 (lung carcinoma), SW620 (colon carcinoma), PANC-1 (pancreas carcinoma), and HuH-7 (hepatocellular carcinoma) were maintained in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10 % fetal bovine serum (FCS), 100 U/ml penicillin, 100 µg/ml streptomycin and 3.5 µg/ml amphotericin B in culture flasks at 37°C with 5 % CO_2 in a humidified incubator. Hela (cervix carcinoma), SK-BR-3, MDA-MB-231 and MCF-7 (mamma carcinoma) were grown in medium Roswell Park Memorial Institute medium (RPMI) with the same supplements as mentioned above.

Cell viability was determined by MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] assays [17]. The appropriate cell numbers were determined beforehand by growth curves. Cells were then seeded onto 96-well plates in octuplicates. After 24 hours, cells were treated with a serial dilution of *Taraxacum* (1 – 200 mg/ml) and cultivated for further 48 hours before the assay was performed.

Cell proliferation was monitored by the xCELLigence technology according to the manufacturers' recommendations. Briefly, the device enables a continuous real-time monitoring of the adhesion properties of cells by detection of variations in electrical impedance that are then transformed to a dimensionless factor referred to as Cell Index [18]. Treatment with serial *Taraxacum* dilutions (0.1 - 200 mg/ml) was conducted 24 hours after seeding of SKOV-3 (2,000 c/well) and NIH:OVCAR-3 (3,000 c/well) cells. The Cell Index was normalized to the time point of *Taraxacum* addition. Growth curves were obtained and compared using RTCA software (Version 1.0).

Cell migration was assessed by "wound healing" experiments using confluent monolayers of cells in six-well microtiter plates. The tip of a micropipette was used to create a linear scratch resulting in two cell fronts, which were ~2 mm apart from each other as described earlier [19, 20]. Cells were either treated with *Taraxacum* (10 mg/ml) or with culture medium as control. Cell migration was judged by photographs taken immediately and at 24 hours as well as 48 hours after scratching using a digital camera and ImageJ software [21]. The test was conducted in four independent experiments.

The invasive potential of ovarian carcinoma cell lines, SKOV-3 and NIH:OVCAR-3 was assessed using Boyden chamber invasion assay. Here, 50,000 cells were seeded in the upper inserts, treated with *Taraxacum* (10 mg/ml) and enabled to migrate along a FCS concentration gradient (5 - 15 %). Subsequently, cells on the bottom of the inserts were fixed and stained with DAPI (4',6-diamidino-2-phenylindole, 1 µg/ml in methanol). Insert-membranes were removed with a scalpel and embedded on a slide using Mowiol 4-88. Fluorescently labeled cells were counted under a fluorescence microscope.

Tumor cells were treated with 10 mg/ml or 100 mg/ml *Taraxacum* extract 24 hours after seeding onto six-well plates. After 24 hours of incubation, cells were harvested by trypsin and fixed with EtOH abs. (-20 °C; 1 d). After RNase A treatment for 45 minutes, DNA was stained with 20 µl PI (propidium iodide; 20 mg/ml), and cell cycle analysis was accomplished using a flow cytometer. Data were processed using the Beckmann coulter CXP software to identify the proportions of each cell population in each cell phase.

For flow cytometric assessment of vital mitochondria, 24 hours after treatment with 10 mg/ml or 100 mg/ml *Taraxacum,* vital mitochondria of SKOV-3 and NIH:OVCAR-3 cells were stained with MitoTracker Deep Red FM (0.1 µM) dye for one hour.

Combinational effects and possible interactions between *Taraxacum* and two different mistletoe extracts (*Iscucin Tiliae* and *Iscucin Pini*), were evaluated using MTT assays. For this purpose, cells were seeded onto 96-well plates and after 24 hours treated with Iscucin (strengths C-G) and *Taraxacum* dilutions (0.1 - 200 mg/ml). Combinatorial effects were analyzed by CompuSyn software based on the method introduced by Chou and Talalay [22, 23] to calculate the combination index (CI). The CI value informs about combinational effects of two or more drugs [24].

The significance of results was analyzed using GraphPad Prism version 5.0. For statistical analysis a Student's *t*-test was carried out. Results are expressed as means and error bars indicate the standard deviation (SD).

Results

Cell viability assays (MTT) were used to evaluate antitumorigenic effects of *Taraxacum* on a panel of cancer cell lines of different origin (Figure 1). The calculated concentration causing half maximal decrease of cell viability (IC_{50}) ranged from 12 - 160 mg/ml. All cell lines derived from gynecological tumors (cervical-carcinoma [Hela], mamma carcinoma [MCF-7, SK-BR-3, MDA-MB-231], endometrial-carcinoma [Ishikawa, AN3 Ca], ovarian carcinoma [NIH:OVCAR-3, SKOV-3]), with an average IC_{50} value of 83 mg/ml were more sensitive to *Taraxacum* than the normal human fibroblast cell line, NHDF-C (IC_{50}: 128 mg/ml). The ovarian carcinoma cell line, NIH:OVCAR-3, was the most sensitive (IC_{50}: 12 mg/ml) and the lung carcinoma, A549, was the most insensitive (IC_{50}: 160 mg/ml) tested cell line. The latter one as well as both urinary bladder carcinoma cell lines (EJ28 and RT112) and colon carcinoma cell line (SW620) were more resistant to *Taraxacum* than the fibroblast cell line. Thus, *Taraxacum* had a cytotoxic effect on most adult tumor cell lines particularly on gynecological tumor cells. Therefore, the present authors chose the ovarian cancer cell lines, NIH:OVCAR-3 and SKOV-3, for further experiments.

Taraxacum with concentrations > 100 mg/ml significantly reduced cell viability in the ovarian cancer cell lines,

A

Tumor entity	Cell line	IC50 [mg/ml]
Ovarian carcinoma	NIH:OVCAR-3	12
Cervical carcinoma	HeLa	40
Mamma carcinoma	SK-BR-3	73
Mamma carcinoma	MDA-MB-231	99
Mamma carcinoma	MCF-7	101
Pancreatic carcinoma	PANC-1	105
Endometrial carcinoma	AN3-CA	109
Ovarian carcinoma	SKOV-3	113
Endometrial carcinoma	Ishikawa	114
Hepatocellular carcinoma	HUH-7	128
Human Fibroblasts	NDHF-C	128
Colon carcinoma	SW620	136
Urinary bladder carcinoma	EJ 28	146
Urinary bladder carcinoma	RT 112	157
Lung carcinoma	A549	160

B

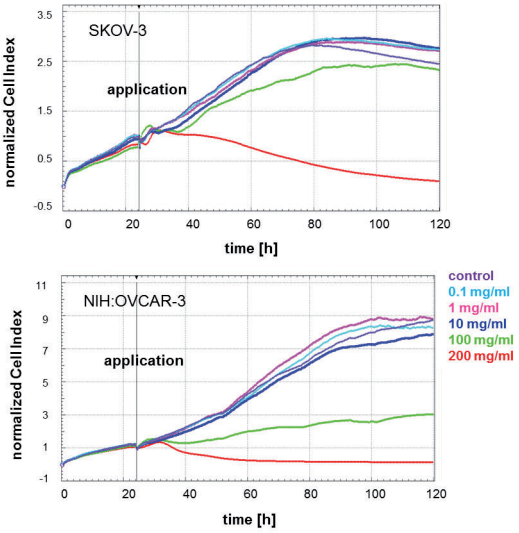

Figure 1. — Anti-proliferative effects of *Taraxacum* extracts on adult tumor cell lines.
A) A panel of 14 adult cancer cell lines is screened for its sensitivity to *Taraxacum* by MTT assays. As control for non-tumorigenic cells, a fibroblast cell line (NHDF-C) is additionally tested. B) Real-time monitoring of cell proliferation by xCELLigence System is performed for ovarian carcinoma cell lines, SKOV-3, and NIH:OVCAR-3, under treatment of 0.1, 1, 10, 100, and 200 mg/ml *Taraxacum*.

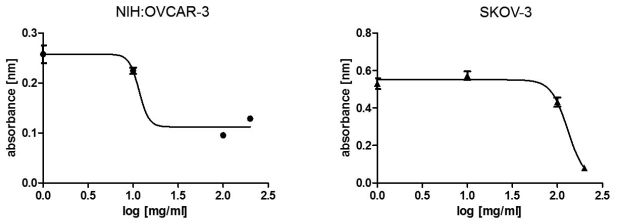

Figure 2. — *Taraxacum* reduces cell viability of ovarian cancer cell lines. MTT-assays (48 hours after treatment) reveal a concentration dependent decrease of cell viability in NIH:OVCAR-3 and SKOV-3 cells.

NIH:OVCAR-3 and SKOV-3, compared to untreated controls. Continuous detection of cell adherence using the Roche xCELLigence device revealed an immediate decline after addition of the highest concentration of *Taraxacum* (200 mg/ml). This decrease in the cell index was correlated with lower cell numbers and consistent with MTT data. Doses of 100 mg/ml caused delayed growth of proliferation in SKOV-3 cells and led to a remarkable growth arrest in NIH:OVCAR-3 cells (Figures 1 and 2).

Besides cell proliferation, migration and invasion are characteristics of aggressive tumor cells. Thus, the present authors performed scratch assays and Boyden Chamber assays to assess the impact of *Taraxacum* on cancer cell migration and invasion, respectively. The results revealed a reduced migration indicated by a delayed closure of the scratch in both ovarian carcinoma cell lines upon *Taraxacum* treatment compared to untreated cells (Figure 3).

Gaps were closed more efficiently in untreated SKOV-3 and NIH:OVCAR-3 cells (78.3 % and 36.5 %) compared to cells with 10 mg/ml *Taraxacum* administration (72.9 % and 31.9 %, 24 hours after scratching). Effects on cell migration became significant ($p = 0.0092$) in SKOV-3 after 48 hours. A complete gap closure was monitored in control dishes, while cells treated with 10 mg/ml *Taraxacum* displayed only 89 % gap closure, respectively. Gap closure was likewise delayed in NIH:OVCAR-3 cells after 48 hours (Figure 3A, B).

The impact of *Taraxacum* on cell invasion was further analyzed by Boyden Chamber assays. A less pronounced effect on invasion compared to migration was observed. Cell invasion seemed to be unaffected in SKOV-3 cells, but an inhibition of cell invasion induced by *Taraxacum* (10 mg/ml) treatment was demonstrated in NIH:OVCAR-3 cells, however, this effect did not reach statistical significance (Figure 3C).

DNA content analyses were performed to study the effects of *Taraxacum* on cell cycle progression and apoptosis induction. Hypodiploid cells, caused by DNA fragmentation in the late stage of apoptosis, could be detected in the subG1-phase of the DNA histogram. An increase of subG1-phase cells could be demonstrated in both ovarian carcinoma cell lines in a dose-dependent fashion 24 hours after treatment (Figure 4A).

Taraxacum (10 mg/ml) increased the fraction of apoptotic cells by 1.6 % and 9.3 % in SKOV-3 and NIH:OVCAR-3 cells, respectively. At higher concentrations of *Taraxacum* (100 mg/ml) a dose-dependent increase in the apoptotic cell fraction was observed (14.7 % in SKOV-3, which was significant, $p = 0.0223$, and 19.4 % in NIH:OVCAR-3 cells, respectively). Other phases of the cell cycle were largely left unaffected by *Taraxacum* exposure. To further analyze apoptotic responses, MitoTracker

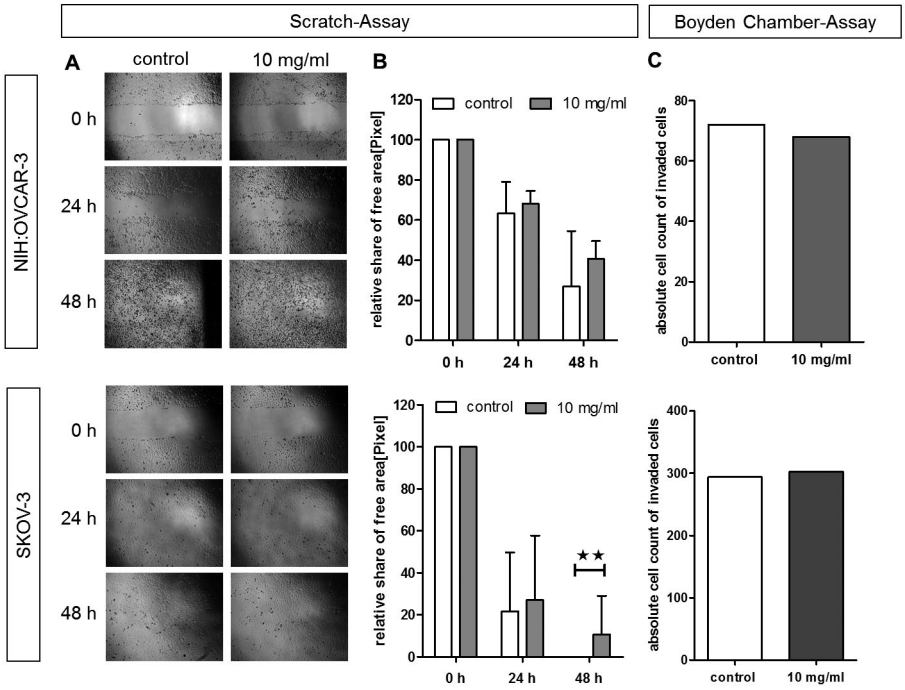

Figure 3. — *Taraxacum* inhibits cell migration and invasion.
A, B) *In vitro* scratch wound healing assays are used to quantify cell migration of SKOV-3 and NIH:OVCAR-3 cells. Scratches are photographed at 24 and 48 hours after treatment of 10 mg/ml (A). Free area of scratch is quantified and normalized to control ($p \leq 0.05$*). B, C) Cell invasion properties after 48 hours *Taraxacum* treatment (10 mg/ml) are assessed via Boyden chamber assays. Invaded cells on the bottom side are DAPI stained and counted under a fluorescence microscope

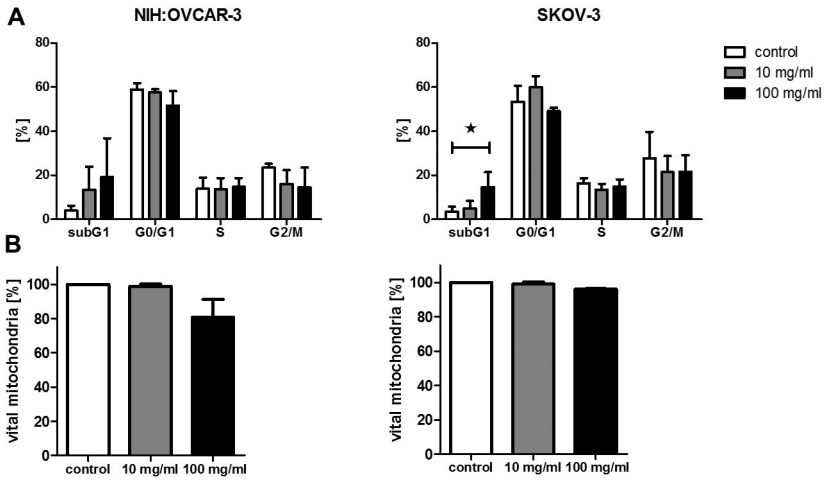

Figure 4. — Taraxacum induced apoptosis and reduced mitochondrial integrity in ovarian carcinoma cells.
A) Cell cycle analysis (PI staining) of ovarian carcinoma cells is performed upon 24 hours Taraxacum treatment (10 mg/ml and 100 mg/ml) ($p \leq 0.05$*). B) Viable mitochondria are stained with Mito Tracker Deep Red FM after 24 hours Taraxacum application (10 mg/ml and 100 mg/ml) and analyzed by FACS. Viable mitochondria fraction is normalized to control cells.

Deep Red staining was performed. Exclusion of this fluorescent dye indicates an impairment of mitochondrial function and an activation of the intrinsic pathway of apoptosis, as it selectively stains vital mitochondria. While 10 mg/ml *Taraxacum* extract had no effect on mitochondrial viability in NIH:OVCAR-3 and SKOV-3 cells, mitochondrial staining was reduced in NIH:OVCAR-3 cells in the presence of 100 mg/ml *Taraxacum* (Figure 4B). In line with the present data on cell viability, SKOV-3 cells were less sensitive to *Taraxacum* treatment in this assay. Taken together, *Taraxacum* extract induced apoptosis and disturbed mitochondrial integrity at high doses in the cell line NIH:OVCAR-3.

MTT assays were performed and CI values were calculated using CompuSyn software to assess combinational effects of *Taraxacum* and two different mistletoe extracts, *Iscucin Pini* or *Iscucin Tiliae,* on cell viability (Figure 5, Table 1). Here, combinations of *Iscucin Pini* or *Iscucin Tiliae* with a high dose of *Taraxacum* (200 mg/ml) caused synergistic anti-proliferative effects as indicated by CI-values <1. The CI fraction of affected cells (Fa) plot also revealed that synergistic effects mainly occurred in highly affected cells. The observed combinatorial effects were

Table 1. – *Taraxacum combined with mistletoe extracts provokes synergistic effects.*

		CI value			
		Iscucin	***Pini Iscucin***	***Pini Iscucin Tiliae***	***Iscucin Tiliae***
***Taraxacum* [mg/ml]**	***Iscucin Pini/ Tiliae* [mg/ml]**	**NIH: OVCAR-3**	**SKOV-3**	**NIH: OVCAR-3**	**SKOV-3**
0.1	$1.56*10^{-5}$	**0.2099**	**0.5743**	1.2134	13.0984
0.1	$3.13*10^{-4}$	4.0046	12.6408	34.174	18.0440
0.1	0.00625	21.4423	29.03480	**0.9608**	1.2479
0.1	0.125	**0.1843**	**0.7856**	**0.6743**	**0.5964**
0.1	2.5	**0.0542**	**0.3359**	2.0272	1.6459
1.0	$1.56*10^{-5}$	2.2351	5.2831	2.1797	4.1128
1.0	$3.13*10^{-4}$	2.1395	7.6702	10.0184	11.2926
1.0	0.00625	5.3935	21.0387	1.1396	1.2002
1.0	0.125	**0.2434**	**0.9403**	**0.5182**	**0.5111**
1.0	2.5	**0.0437**	**0.2344**	**0.8392**	**0.7209**
10.0	$1.56*10^{-5}$	3.3705	24.1363	6.5312	outlier
10.0	$3.13*10^{-4}$	3.6317	25.0250	8.4433	25.4133
10.0	0.00625	4.4454	25.8860	1.3321	1.5280
10.0	0.125	**0.3153**	1.4101	**0.6040**	**0.6801**
10.0	2.5	**0.0659**	**0.4902**	1.3981	1.3926
100.0	$1.56*10^{-5}$	6.3948	14.4195	outlier	9.3587
100.0	$3.13*10^{-4}$	6.0367	13.8388	8.8052	8.1076
100.0	0.00625	5.8205	10.5542	1.5919	1.9578
100.0	0.125	**0.6690**	1.9537	**0.5560**	1.0244
100.0	2.5	**0.1647**	1.1534	1.1813	**0.8421**
200.0	$1.56*10^{-5}$	**0.1670**	**0.3287**	**0.2324**	**0.2672**
200.0	$3.13*10^{-4}$	**0.1615**	**0.2956**	**0.2375**	**0.2145**
200.0	0.00625	**0.2832**	**0.3150**	**0.5417**	**0.1867**
200.0	0.125	**0.3867**	**0.2959**	1.0233	**0.5499**
200.0	2.5	**0.2610**	1.1597	2.1315	2.1020
	Median	**0.387**	**1.682**	**1.197**	**1.320**

CI values < 1 (synergistic effects) are in grey; CI values >1 indicate antagonistic effects.

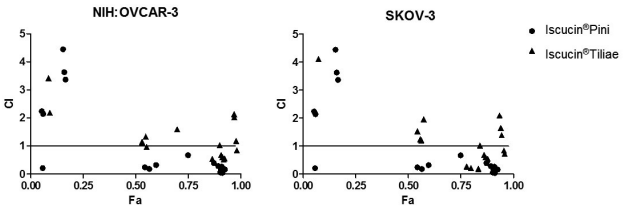

Figure 5. — Taraxacum combined with mistletoe extracts provokes synergistic effects. Combination indices (CI) are plotted against the fraction of affected cells (Fa; 0.05-0.097) to analyze synergistic effects of simultaneous treatment of Taraxacum and *Iscucin Pini* or *Iscucin Tiliae*. CI values were calculated by CompuSyn (http://www.combosyn.com) and CI values < 1 indicate a synergistic, CI value = 1 indicates an additive, and CI values > 1 indicate antagonistic effects.

similar for both mistletoe extracts indicating specific effects.

Discussion

Ovarian cancer is the most lethal gynecologic neoplasm and is still difficult to cure [25, 26]. Standard treatment includes taxane- (paclitaxel or docetaxel) and platinum (cisplatin or carboplatin) chemotherapy. These kinds of therapies are often associated with neurotoxicity and ototoxicity, as well as drastic adverse effects such as nausea, vomiting, as well as thrombocytopenia and leucopenia [27, 28]. Therefore, patients suffering from ovarian cancer frequently use CAM as supportive therapies with the intention to reduce adverse effects of other medications, to improve quality of life, and to assist in curing their disease. Mistletoe preparations, representing one of the most popular CAM therapies in oncology, have already been used for cancer patients. Iscador, which has been evaluated in numerous clinical trials, had a positive effect on overall survival and well-being for ovarian cancer patients [29]. However, systematic analyses and Cochrane reviews have carved out the need for more rigid trial designs to evaluate the impact of mistletoe preparations on patient survival [30, 31]

Recent studies also postulate anti-tumoral effects of *Taraxacum officinale* plant extracts on diverse tumor entities including ovarian carcinoma [32, 33–36]. Here, the present authors investigated the potential anticancer properties of an aqueous fermented *Taraxacum* extract, *Taraxacum e planta tota ferm 34c,* alone, and in combination with frequently administered aqueous mistletoe extracts. Among all tested adult tumor entities, gynecological tumors

(IC_{50}: 83 mg/ml), including ovarian-, cervical-, and mamma carcinoma, were most susceptible to *Taraxacum* exposure and more sensitive than the human fibroblast cells, NHDF-C (IC_{50}: 128 mg/ml). This result indicates that cytotoxic effects of *Taraxacum* are cancer cell specific. In particular, the ovarian carcinoma cell line, NIH:OVCAR-3, was the most sensitive cell line, supporting previous results that *Taraxacum* extracts are a promising supportive therapy option for this tumor entity [29].

In comparison to IC_{50} values determined for breast cancer stem cells in a 3D-spheroid test system (approximately 1 mg/ml) for ethanolic and methanolic extracts [37], the present IC_{50} values were comparably high. This underlines that the extraction process and the used solvent plays a crucial role in the effectiveness of the extracts. Moreover, the fermentation, which modifies the substances post extraction, will affect the resulting composition. Thus, direct comparison of anti-proliferative effect of extracts originating from diverse extraction methods requires careful interpretation. In the present series of experiments, induction of apoptosis, marked by an enlargement of the subG1 peak in cell cycle analyses and by an impairment of mitochondrial function was most prominent in the ovarian carcinoma cell line NIH-OVCAR:3. These results are in line with triggering of the intrinsic apoptotic pathway induced by *Taraxacum* treatment. Similar results have been reported by Chatterjee *et al.* in the human melanoma cell line A375. Here, the authors found an induction of apoptosis and a dissipation of mitochondrial membrane potential using aqueous *Taraxacum* root extract [15].

As the prevention of metastases could be a promising attempt in combating ovarian cancer, the effects on cell migration and invasion were investigated. In the scratch assay for *in vitro* directed migration, the present authors demonstrated a reduced migratory capacity in both ovarian carcinoma cell lines investigated. However, decreased invasiveness could only be demonstrated in NIH:OVCAR-3 cells. This cell line is characterized as weakly invasive, whereas SKOV-3 cells are strongly invasive [38]. In line with the present results, aqueous extracts from *Taraxacum officinale* showed to affect signaling pathways (FAC/SRC) and proteins (matrix-metalloproteinases) involved in invasion and migration processes [12].

CAM-treated cancer patients sometimes receive a combination therapy comprising *Taraxacum* and mistletoe extracts. In order to analyse synergistic or even antagonistic effects of this co-treatment, CI values were calculated. Apparently, some drug combinations in the present study were detected as synergistic (CI < 1), others as antagonistic (CI > 1), but observed effects were similar for tested cell lines and both mistletoe extracts indicating specific effects. Synergistic effects particularly occurred when cells were highly affected (high Fa-values). Results underline the requirement to study combination treatments to define appropriate concentrations evoking synergistic effects and to elucidate underlying pathways as a basis for transfer into clinical practice.

Conclusion

The present authors identified anti-proliferative, apoptosis-inducing, as well as inhibitory migrative and invasive effects of an aqueous fermented *Taraxacum* extract on ovarian carcinoma cell lines. In addition, at certain concentrations, *Taraxacum* and mistletoe extracts revealed synergistic effects. Despite these promising results, further research is required to characterize the active substances from *Taraxacum*, responsible for its anticancer effects and to determine combinational effects between *Taraxcacum* and conventional chemo- or radiotherapy.

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