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Review paper

Interferon and hormone sensitivity of endocrine-related tumors

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Interferons (IFNs) have been shown to enhance both *in vitro* and *in vivo* the antiproliferative activity of some hormones and anti-hormones which mainly act via steroid receptors. We discuss some of the mechanisms which could be involved in determining this effect in breast, endometrial and prostatic cancer cells, with a particular emphasis on steroid receptor modulation, reduction of the expression of epidermal growth factor receptors and, finally, down-regulation of some oncogenes. It seems that under appropriate conditions IFN might produce changes in cancer cells that enhance or restore hormone sensitivity. Nevertheless, available clinical data are too few to allow any conclusion to be drawn and this problem merits further investigations.

Key words: Hormone, interferon, resistance, sensitivity.

Introduction

Interferons (IFNs) have limited activity in cancer treatment. Nevertheless, they are known to possess antiproliferative, differentiative and immunomodulatory activities, probably exerted through separate mechanisms.[1] A growing body of evidence from laboratory research now supports the concept that IFNs can complement *in vitro* the activity of some hormones and anti-hormones currently used in breast cancer therapy.[2–11] Further, there are some clinical data suggesting that the combination IFNs/tamoxifen (TAM) can improve the response to TAM alone or overcome the resistance to the antiestrogen in metastatic breast cancer patients.[12–18]

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Biological bases of the complementation of hormone and antihormone activity by IFNs

It has been shown that simultaneous exposure to IFNs and TAM or IFNs and medroxyprogesterone acetate (MPA) can result in enhanced antiproliferative effects compared with either single agent alone in human estrogen-sensitive mammary cancer cells, containing estrogen and progesterone receptors (ER and PR).[2–5,7–10] The same result can be obtained if cultures are pretreated with IFNs and subsequently exposed to TAM or MPA[4,5,7,11] (see also Tables 1–3). In this case, the promotion of the antitumor effect of the above-mentioned drugs can be achieved with very low concentrations of IFN. The combination treatment has been reported to be more effective, if compared with single agents, even *in vivo*. when ER positive and estrogen-sensitive cells (ZR-75-1 or MCF-7) are inoculated in nude mice.[19,20] The synergistic/additive activities of combinations of IFNs and TAM may be the result of different mechanisms.

A series of studies indicated that both natural and recombinant IFNs are able to increase the steroid hormone receptor level *in vitro* and *in vivo* (Tables 1–4). In 1982, Pouillart *et al.* showed that the administration of fibroblast IFN (nIFN-β) (8 i.m. injections of 6×10^6 IU over a period of 40 days) to six patients with metastatic breast cancer determined an increase in ER and PR.[21] In particular ER was enhanced in tumor biopsies of two out of two and PR in five out of six patients tested, respectively. Subsequently, other authors suggested IFN-induced enhancement in steroid receptors in different *in vitro* systems, such as human breast cancer cells, human endometrial explants, human endometrial cancer cell lines and leukemic cells.[5,8–11,22,25] In this context, Sica *et al.* showed that both nIFN-β and recombinant α2b IFN (rIFN-α2b) increase ER and PR, at concentrations ranging from 10 to

Table 1. Summarized data from the literature concerning the effects of IFN-α/TAM on cell proliferation and IFN-α on ER levels in breast cancer cell lines

IFN/cells		Dose of IFN (IU/ml)	Time of treatment[a] (days)	ER (% of control)	Time of treatment[b] (days)	Cell no. (% of control)	Combination of IFN with TAM (growth inhibition)	Reference no.
rIFN-α								
BT-20	(ER^-)	500	–	–	14	77	>TAM=IFN	2
CG-5	(ER^+)	500	6	91	6	85	<TAM>IFN	3
MCF-7	(ER^+)	111–6000	5	100		–	–	27
ZR-75-1	(ER^+)							5
low density		10	2	308	6	90	>TAM>IFN	5
high density		10	2	100	–	–	–	5
MCF-7	(ER^+)	10	5	100	5	97	=TAM>IFN	6
		100	5	106	5	86	=TAM>IFN	6
		1000	5	109	5	70	>TAM>IFN	6
MCF-7	(ER^+)	500	3	130	3	50	>TAM>IFN	11
		1000	3	150	3	32	>TAM>IFN	11
CG-5	(ER^+)	10	5	117	6	78	>TAM>IFN	9
		100	5	141	6	77	>TAM>IFN	9
		1000	5	143	6	51	>TAM>IFN	9
IFN-α								
ZR-71-1	(ER^+)	500	4	40	9	64	–	37

[a]Time of treatment before ER determination. [b]Time of treatment before the evaluation of cell growth.

Table 2. Summarized data from the literature concerning the effects of IFN-β/TAM on cell proliferation and IFN-β on ER levels in breast cancer cell lines

IFN/cells		Dose of IFN (IU/ml)	Time of treatment[a] (days)	ER (% of control)	Time of treatment (days)	Cell no. (% of control)	Combination of IFN with TAM (growth inhibition)	Reference no.
nIFN-β								
CG-5	(ER^+)	500	6	63	6	54	>TAM>IFN	3
CG-5	(ER^+)	10	–	–	6	–	>TAM>IFN	4
CG-5	(ER^+)	10	5	171	6	90	–	22
MCF-7	(ER^+)	10	5	111	5	88	=TAM>IFN	6
		100	5	119	5	59	>TAM>IFN	6
		1000	5	–	5	11	>TAM>IFN	6
rIFN-β_{ser}								
MCF-7	(ER^+)	100–200	2	100	7	68	>TAM>IFN	7
T47D	(ER^+)	100	–	–	7	79	>TAM>IFN	7

[a] Time of treatment before ER determination. [b] Time of treatment before the evaluation of cell growth.

Table 3. Summarized data from the literature concerning the effects of IFN-γ/TAM on cell proliferation and IFN-γ on ER levels in breast cancer cell lines

IFN/Cells		Dose of IFN (IU/ml)	Time of treatment[a] (days)	ER (% of control)	Time of treatment[b] (days)	cell no. (% of control)	Combination of IFN with TAM (growth inhibition)	Reference no.
rIFN-γ								
BT-20	(ER^-)	500	4	100	14	77	>TAM=IFN	2
ZR-75-1	ER^+)	500	4	100	14	55	>TAM=IFN	2
MCF-7	(ER^+)	500	–	–	14	75	>TAM>IFN	2
CG-5	(ER^+)	500	6	65	6	61	>TAM>IFN	3
MCF-7	(ER^+)	100	5	104	5	99	=TAM>IFN	6
			5	105	5	88	>TAM>IFN	6
			5	103	5	88	>TAM>IFN	6
ZR-75-1	(ER^+)	100	2	130	7	96	>TAM>IFN	8
MCF-7	(ER^+)	10	–	–	2	90	>TAM>IFN	10
		100	2	128	2	66	>TAM>IFN	10
		1000	–	–	2	70	>TAM>IFN	10
ZR-75-1	(ER^+)	10	–	–	2	74	>TAM>IFN	10
		100	2	165	2	56	>TAM>IFN	10
		1000	–	–	2	48	>TAM>IFN	10
MCF-7	(ER^+)	50/100	2	74	7	74	>TAM>IFN	7

[a] Time of treatment before ER determination. [b] Time of treatment before the evaluation of cell growth.

Table 4. Summarized data from the literature concerning the effects of IFN on ER and PR levels in patients affected by breast cancer

IFN	Dose of IFN (IU)	Patients (no.)	Increase of ER (no.)	Increase of PR (no.)	Side effects	Reference no.
nIFN-β	6×10^6 every 5 days/6 weeks	11 (M)	2/2	5/6	mild	21
rIFN-α	3×10^6 m^2 daily/2 weeks	2/7 (P/M)	3/5	1/5	–	12
nIFN-β	4×10^6 3 times a week/1 week	12 (P)	7/12	4/10	–	32
nIFN-β	2 or 6×10^6 3 times a week/2 weeks	45 (M)	23/38	20/36	mild	28
rIFN-α2b	3×10^6 3 times a week	7 (M)	5/7	1/7	mild	16

(P) = primary cancer; (M) = metastatic cancer.

and PR, at concentrations ranging from 10 to 1000 IU/ml of culture medium, in human mammary estrogen-sensitive CG-5 cells and this is consistent with the increase of ER mRNA observed in the same model after nIFN-β treatment.[9,22,26] Moreover, Bezwoda and co-workers reported that the maximum IFN-induced increase in ER level coincides with the most marked synergistic effect of rIFN-α2b plus TAM in MCF-7 cells.[11]

Biochemical characteristics of steroid receptor molecules newly synthesized under IFN action are essentially identical with the original ones.[5,9,10,19,24]

Some authors, on the other hand, reported no effect or decreased receptor content in response to IFN treatment in breast cancer cells. These conflicting data are probably due to the different types of IFNs, cell lines, culture conditions and steroid receptor assays used.[2,3,6,7,20,27]

More recently, in a phase II randomized study, Sica *et al.* found that *in vivo* treatment with two different doses of nIFN-β (2×10^6 or 6×10^6 IU i.m. three times a week) produced an increase in ER and PR levels in a high percentage (more than 50%) of 38 postmenopausal patients who underwent a biopsy of skin metastases before and after nIFN-β administration. In addition, linear regression analysis of the data indicated that, while there was no correlation between ER and PR content before IFN treatment, this correlation appeared at the end of the IFN course. These findings suggest that IFN exerts a regulatory action on the steroid receptor mechanism, influencing first of all ER and determining via ER a PR enhancement.[28]

Similar information derives from another study performed by the same research group on patients affected by primary endometrial cancer who were randomized to be treated with two different doses of nIFN-β (2×10^6 or 6×10^6 IU i.m. three times a week for 1 week) (see Table 5). Even in this case, nIFN-β modified steroid hormone receptor content which was evaluated in a sample taken under hysteroscopic control, before IFN treatment, and on a specimen obtained at surgery from the same area of the first biopsy, after IFN administration. In particular, ER and PR were increased in 65 and 72%, respectively, of patients who received the lower dose of nIFN-β and in 72 and 60% of patients who were treated with the higher dose of nIFN-β. Moreover, in many cases, the enhancement was higher than 100 fmol/mg protein.[29]

PR is generally regarded as one of the proteins synthesized in response to the activation of ER, so the effect of IFN on PR suggested ER to be true with the native functions. In both *in vivo* studies the affinity of ER and PR after IFN treatment was substantially unmodified with respect to that found before administration of the drug. This confirms what was observed *in vitro*.

The above-mentioned findings concerning the effect of nIFN-β on receptors in endometrial cancer are consistent with results obtained by Scambia *et al.*, who observed an increase of ER and PR in a smaller number of patients affected by primary endometrial cancer who were treated with rIFN-α2b at 5×10^6 IU/day for 5 days.[30] In this study a concomitant decrease in receptors for epidermal growth factor (EGF-R) was observed. On the contrary, a previous paper by Kauppila *et al.* failed to demonstrate any changes of ER and PR levels (evaluated on the 24th day of the menstrual cycle on normal endometrial tissue) after s.c. administration of 3×10^6 IU of IFN-α/day from day 3 to day 23 of the menstrual cycle to five normally cycling healthy women. Nevertheless, in this case, receptor values were expressed relative to those measured from the samples taken on the same day during the preceding control cycle, which is questionable.[31]

Modulation of steroid receptors in endometrial cancer *in vivo* is supported by the findings of Angioli *et al.*, who demonstrated that four different IFNs determine a significant enhancement of PR in AE-7 human endometrial cancer cells cultured *in vitro*, IFN-β being more rapid in the induction of the effect.[25]

Finally, IFN has been shown to enhance ER and PR levels in primary breast cancer also, as reported by Marchetti *et al.*[32] and Hakes *et al.*[12]

The increased expression of steroid hormone receptors may render neoplastic cells more sensitive to the action of drugs mainly acting via receptors themselves. In fact, it is well-known that receptor content is related to response to hormono-therapy in both mammary and endometrial cancer.[33–35]

When IFN and TAM or MPA are co-administered, the influence of the antiestrogen or the progestin on IFN receptor (IFN-R) expression cannot be neglected. TAM slightly increases and MPA significantly enhances IFN-R in ZR-75-1 cells, while estradiol leads to a consistent reduction of IFN-R molecules.[36] Thus IFN-R down-regulation could be prevented by the association of IFN with TAM or MPA, IFN action being facilitated.

Another important finding concerning the biological effects of IFNs, which could be involved in the promotion of the antitumor effect of antiestrogens, is represented by the induction of the synthesis of inhibitory growth factors. IFN-α has been reported

Table 5. Summarized data from the literature concerning the effects of IFN on endometrial ER and PR expression

IFN	Dose of IFN (IU)	Patients (no.)	Tissue	Increase of ER (no.)	Increase of PR (no.)	Side effects	Reference no.
IFN-α	3×10^6/day/from 3rd to 23rd day of the menstrual cycle	5	normal	no change	no change	mild	31
rIFN-α2b	5×10^6/day/5 days	13	primary cancer	9/13	10/13	–	30
nIFN-β	2×10^6 3 times a week/1 week	20	primary cancer	13/20	13/18	mild	29
	6×10^6 3 times a week/1 week	20	primary cancer	13/18	12/20	mild	29

Table 6. Summarized data from the literature concerning the effects of IFN/TAM treatment in patients affected by breast cancer

IFN	Dose of IFN (IU)	Dose of TAM (mg/day)	Patients (no.)		Side effects	Overall response	Stabilization of disease	Reference no.
rIFN-α2c	2×10^6/daily	30	3	(M)	severe		1/3	6
rIFN-α2b	$3 \times 10^6/m^2$ daily/2 weeks	10	2/7	(P/M)	–	1/9	–	12
rIFN-α2b	$5 \times 10^6/m^2$ daily/1 week	20	10/23(A)	(M)	severe	0/9	3/9	59
			13/23(B)		severe	5/10	3/10	59
nIFN-α	5×10^6 every 2 days	30	13	(M)	severe	2/13	11/13	13
nIFN-β	3×10^6/day/2 weeks	30[a]	33/43(C)	(M)	mild	8/33	16/33	14
			10/43(D)		mild	3/10	3/10	14
nIFN-β	1×10^6 3 times a week	30	23	(M)	mild	2/22	18/22	15
nIFN-β	2 or 6×10^6 3 times a week/2 weeks	30[a]	45	(M)	mild	9/23	7/23	22
rIFN-α2b	3×10^6 3 times a week	20[a]	7	(M)	mild	4/7	–	16

(A) prior TAM exposure; (B) no prior TAM exposure; (C) progressive to TAM; (D) stable or partially responsive to TAM. [a]Starting from day 15. (P) = primary cancer; (M) = metastatic cancer.

mRNA levels in ZR-75-1 cells.[37] On the other hand, previous studies have indicated that TAM does induce the secretion of TGF-β in MCF-7 cells.[38] The antiproliferative response to both the antiestrogen and IFN-α can be blocked by co-treating the cells with TGF-β antibodies.[37,38]

It can by hypothesized that in the development of the enhanced antiproliferative effect which occurs when IFNs plus TAM are used, the interference with the autocrine growth loop that controls estrogen-sensitive cell proliferation may have an important role.[37,38]

Changes in EGF-R were observed concomitant with the inhibition of cell proliferation in CG-5 cells treated with IFN. If exposed to nIFN-β for 24 h, CG-5 cells seem to show an increase in EGF-R, but when the treatment is prolonged from 72 to 120 h a reduction of receptors is observed with respect to control with the highest concentration used (1000 IU/ml of culture medium). rIFN-α2b requires a longer time of action to produce the same effects (Iacopino *et al.*, manuscript submitted).

These findings are in agreement with data from Chakravarthy and co-workers concerning MDA468 cells.[39] In addition, IFN-α-induced down-regulation of EGF-R was also shown by Eisenkraft *et al.*[40] in renal carcinoma cells and by Zoon *et al.*[41] in bovine kidney cells.

More interesting are the above-mentioned data concerning the decrease in EGF-R with a concomitant increase in ER and PR in patients affected by primary endometrial cancer treated with rIFN-α2b.[30]

Variations of EGF-R due to IFN could be of relevance in view of the inverse relationship between the presence of ER, PR and EGF-R in human breast and endometrial tumors, the decrease in EGF-R being associated with a higher differentiation of neoplastic cells and a better clinical prognosis.[42–44]

Very recently, Sica *et al.* observed that CG-5 cells treated with 100–1000 IU/ml of recombinant IFN-β showed a relevant decrease in c-*myc* and c-*erb* B2 expression analyzed by Western Blot technique. The effect of 1000 IU/ml was observed starting from 48 to 72 h of treatment; the inhibition of oncogene protein expression reached about 50% with respect to control at 120 h and persisted for both c-*myc* and c-*erb* B2 after 168 h (Sica *et al.*, manuscript submitted). The decrease in oncoproteins seems to be due to an indirect effect of IFN and it is not strictly linked to the antiproliferative effect of the drug, because it occurs in parallel with the maximal growth inhibition observed. It could be important in view of previous observations of Russel *et al.* who showed that the activation of the ER mechanism determines a repression of the transcription of the *neu*-protooncogene (HER-2 or c-*erb* B2).[45]

Thus in CG-5 cells IFN seems, on the one hand, to increase steroid hormone receptors (which are considered a sign of differentiation) and, on the other hand, decrease oncogene expression (which is related to a particular aggressiveness of the tumor and to a poor prognosis). Moreover, it has to be kept in mind that the c-*erb* B2 oncogene encodes a protein which has structural and sequence similarity with the EGF-R.[46]

Finally, it is known that IFNs and TAM share the capability of increasing *in vivo* natural killer activity.[47–49] It can be discussed if the clinical effect of the two drug combination is due to its immunological activity.

Clinical data concerning the association of IFN/TAM or IFN/MPA in breast cancer

IFN-β plus TAM

An Italian study by Buzzi *et al.* showed that the association of TAM/IFN-β is able to improve the responsiveness in hormone-sensitive patients and to restore the hormone sensitivity in patients who became resistant. These authors, in a phase II trial, enrolled 43 patients affected by advanced breast cancer progressive (group A) and stable or partially responsive (group B) to previous treatment with TAM. These patients were treated for 14 days with nIFN-β (3×10^6 IU/day) and subsequently exposed to TAM (30 mg/day) and nIFN-β (at the same dose used before) once a week. Patient receptor status was positive or unknown. The overall response rate was 26% with eight partial responses obtained in group A and three complete responses in group B. Stabilization of disease was observed in 44% of cases. Toxicity was mild,[14] Table 6.

In agreement with these observations, Cartei *et al.* showed two partial responses and 18 stable diseases in 23 patients with metastatic breast cancer in progression during TAM 30 mg/day, adding 3×10^6 IU/day of nIFN-β three times a week,[15] Table 6.

Repetto *et al.* used the association of IFN-β/TAM in 39 metastatic breast cancer patients as follows: nIFN-β 6×10^6 IU s.c. 3 times a week for two weeks, followed by nIFN-β 3×10^6 IU every other day plus TAM 20 mg/day. Twenty-eight patients with measurable disease were evaluable for the response. The authors observed one complete response and six partial responses. Stable disease was obtained in 46.4% of cases and progression in

28.5%. Also in this study toxicity was mild.[17] The authors conclude that on the basis of their findings and of data from other similar studies, as the combination of IFN-β and TAM has an established *in vitro* rationale, a randomized trial comparing such a combination with TAM appears justified.

A combination therapy of IFN-β plus IFN-γ plus TAM was used by Peretz *et al.* in 16 ER-negative breast cancer patients, with an objective response in 38% of cases.[18] This suggests that the association of two types of IFN could be effective in inducing a more differentiated phenotype in cells which have lost estrogen dependence (ER-negative) and behave more aggressively. The clinical response obtained by Peretz and co-workers could be explained on the basis of the activation of the mechanism of action of IFN by TAM. In fact, Lindner *et al.* observed, *in vitro*, that cells treated with TAM and subsequently exposed to IFN-β showed an enhanced expression of IFN-sensitive genes (protein kinase R and 2′5′oligoadenylate synthetase). Fold enhancement was greater in ER-negative BT-20 cells compared with ER-positive MCF-7 cells. The ability of modulating IFN sensitive genes in cells that lack ER supports a non-classical mechanism of action.[50] Moreover, ER-negative human breast cancer cells, as well as ER-positive cells, are sensitive to the action of negative growth factors, such as TGF-β,[38,52] which are induced by both IFN and TAM.[37,38]

The combination of two types of IFN, used by Petetz *et al*, can be justified by results obtained *in vitro* by Gastl *et al.* In fact, these authors demonstrated, in 1985, that the combination of rIFN-γ and rIFN-α2 increases HLA-DR expression, which is a sign of differentiation. They suggested that this association might be useful in the treatment of breast cancer, providing a higher cytostatic effect than single agent alone in both ER-negative and -positive human breast cancer cell lines and modulating cell membrane properties.[52]

Recchia *et al.*[53] treated 49 patients, affected by metastatic inoperable breast cancer with evidence of progressive disease, with a combination of nIFN-β, TAM and retinyl palmitate. Among the evaluable patients, 55% achieved a clinical response, 20% had a stable disease and 25% progressed. Median time to failure was 23.6 months. Median overall survival was 19 months. Toxicity was moderate. In this case retinoids, which are known *per se* to have a differentiative effect,[54] were added to nIFN-β and TAM. The combination could be justified by the evidence that retinoids have been reported to dramatically increase the antiproliferative action of IFN in ER-positive and ER-negative breast cancer cells.[55,56] Moreover, Lama *et al.* showed that nIFN-β and retinoids synergize in reducing estrogen-sensitive CG-5 breast cancer cell growth[57] and have a subadditive effect in estrogen-insensitive cells (manuscript in preparation). Finally, retinoids have been shown to increase ER in estrogen-sensitive MCF-7 cells and in some sublines resistant to the antiestrogen TAM.[58]

IFN-α plus TAM

Hakes *et al.*, besides demonstrating an increase of steroid hormone receptors in patients affected by disseminated breast neoplasia, found that one of them, progressive after three prior hormone trials including TAM, when continued on the combination TAM/rIFN-α2b achieved a partial response lasting 5 months.[12]

In line with these data, Seymour *et al.* obtained one complete and three partial responses in four of seven patients treated with 3×10^6 IU rIFN-α three times per week for 2 weeks and 20 mg TAM per day by month, starting from the 14th day.[16]

Other authors tried the combination IFN/TAM (see Table 6); nevertheless, on the basis of the results obtained with the addition of IFN-α to TAM, it seems that in the majority of cases no therapeutic benefit was demonstrated. Some authors had to discontinue the treatment or reduce the doses of IFN administration because of the toxicity, which was linked to the high dosage of drug administered (5×10^6 IU/m^2 i.m. for 5 days each week or 5×10^6 IU every 2 days s.c.).[13,59]

It should be pointed out that the percentage of stabilization of disease was remarkable in the studies performed with IFN-α as well as in those in which IFN-β was used (see Table 6).

IFNs plus MPA

Very few data are reported regarding the association of IFN and MPA in breast cancer patients. Wildfang *et al.* observed one out of nine complete responses and two out of nine partial responses in a phase II trial with a combination of 1000 mg/day MPA plus 5×10^6 IU of IFN, given s.c. three times per week, in patients with ER/PR-positive advanced breast cancer and progressive under hormonal treatment.[60] Similar results are reported by Fedeli *et al.*[61] Few patients were included in both these studies, which does not allow us to draw any conclusions.

Nevertheless, it is to be pointed out that also in these studies toxicity was very mild.

Effects of IFN on prostatic cancer cells

A limited number of *in vitro* studies have been published on the effects of IFNs in prostate cancer cells compared with those related to breast cancer cells.

Data from our laboratory show that nIFN-β inhibits cell growth of PC-3 cells, a human prostatic adenocarcinoma cell line, which is hormone-insensitive.[62,63] In addition, it produces an enhancement of androgen receptor (AR) levels, evaluated by a whole cell assay in PC-3 cells. This increase seems not to be related to a selective block of PC-3 cells in any phase of the cell cycle. Pretreatment with nIFN-β determines a partial responsiveness of PC-3 cells to dihydrotestosterone and to the antiandrogen hydroxyflutamide.[64]

Very recently, preliminary experiments by Sica *et al.*, showed that nIFN-β causes a reduction of c-*myc* mRNA in PC-3 cells treated for 2–4 h with the drug,[65] in agreement with results obtained by the same authors in breast cancer cells, where the decrease of oncoproteins was found after a longer time of treatment.

Other mechanisms involved in the antiproliferative and/or differentiative effect of IFN in prostatic cancer cells have been proposed by different authors.

Okatami *et al.* studied the correlation between changes in intracellular cAMP level and IFN.[66] cAMP is a regulator of cell metabolism, growth and differentiation,[67–69] and the cAMP level is increased in PC-3 cells treated with IFN.[66] Moreover, it has been reported that cAMP can restore the integrity of normal negative growth regulatory pathways that have been disrupted in the process of malignant transformation.[69–72] One of these mechanisms can be represented by TGF-β_2 secretion, which in PC-3 cells is induced by c AMP.[73] TGF-β is highly growth inhibitory to PC-3 cells.[73,74]

Blumenfeld *et al.* showed that IFN-γ induces HLA-DR expression on the DU-145 prostate carcinoma cell line, raising the theoretical possibility that malignant prostatic cells may be induced *in vitro* to express HLA-DR (malignant prostatic epithelium does not express HLA-DR and it is rarely, if ever, infiltrated by lymphocytes) and become susceptible to immune regulation.[75]

These results are in line with the above findings concerning breast cancer cells and indicate that the differentiative activity of IFNs occurs in a variety of tumors cells of different origin.

Clinical data on IFN alone or IFN associated with anti-androgen in prostatic cancer

From the clinical point of view, carcinoma of the prostate has been only superficially explored as to its susceptibility to IFN without any apparent success. Limited clinical trials of IFNs in the treatment of prostatic cancer were conducted.

Chang *et al.* evaluated the usefulness of rIFN-α2 in nine patients with advanced carcinoma of the prostate, but failed to observe any beneficial effect; this indicates that IFNs are ineffective in arresting the growth of metastases in this stage.[76] Similar disappointing results are also reported by Bulbul *et al.* who treated 16 patients with IFN-β.[77] On the contrary, van Haelst-Pisani *et al.* observed in two patients with extraosseous disease treated with rIFN-α no response in bone metastases, but complete and partial regression of nodal disease.[78] In these studies were enrolled patients with hormone-resistant prostate carcinoma, IFN was administered as single therapy and in both trials with rIFN-α toxicity was substantial.

The possibility to use IFNs to potentiate or restore the hormone sensitivity in prostatic cancer seems more interesting as compared with the use of the drug as single agent. Voce *et al.* tested the association of nIFN-β with antiandrogenic therapy in 15 patients affected by advanced prostatic cancer, who were progressive under antiandrogen therapy. Patients were treated with 3×10^6 IU nIFN-β three times a week and continued antiandrogen therapy. No complete objective response with tumor mass reduction was observed. Nevertheless, patients had a longer survival and a better performance status.[79]

Polledro *et al.* administered intralesionally 2000 IU nIFN-β per day for 5 days once per month for 3 months in 20 progressive patients with prostatic cancer. During this period antiandrogenic therapy was interrupted and it was restarted at the end of IFN administration. Local and general conditions were ameliorated in 25% out of 15 evaluable patients and 30% of them showed stabilization of disease. This suggests that the use of nIFN-β can give good short-term results.[80]

All the authors agree upon the need of further clinical trials with other treatment schedules and dosages of IFNs to determine the IFN role in the treatment of prostatic cancer.

Conclusion

On the basis of the presented data, it seems that, under appropriate conditions, IFN is able to induce changes in tumor cells which could enhance or induce hormone sensitivity both *in vivo* and *in vitro*. This could be of particular relevance from the clinical point of view in patients with primary or acquired resistance to endocrine therapy.

Available clinical data are too few to allow any conclusion to be drawn, but information derived from these studies should provide a stimulus for further research as the interactions among hormone receptors status, growth factors and growth factor receptors, oncogenes, endocrine therapy and role of IFNs are complex and remain to be defined.

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