

AD \_\_\_\_\_

Award Number: DAMD17-00-1-0313

TITLE: New Agents for Taxol-Resistant Breast Adenocarcinoma

PRINCIPAL INVESTIGATOR: Jim Klostergaard, M.D., Ph.D.

CONTRACTING ORGANIZATION: The University of Texas  
M.D. Anderson Cancer Center  
Houston, Texas 77030

REPORT DATE: July 2003

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command  
Fort Detrick, Maryland 21702-5012

DISTRIBUTION STATEMENT: Approved for Public Release;  
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.

20040224 027

# REPORT DOCUMENTATION PAGE

Form Approved  
OMB No. 074-0188

Public reporting burden for this collection of information is estimated to average 1 hour per response, including the time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing this collection of information. Send comments regarding this burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Washington Headquarters Services, Directorate for Information Operations and Reports, 1215 Jefferson Davis Highway, Suite 1204, Arlington, VA 22202-4302, and to the Office of Management and Budget, Paperwork Reduction Project (0704-0188), Washington, DC 20503

<b>1. AGENCY USE ONLY</b> (Leave blank)		<b>2. REPORT DATE</b> July 2003	<b>3. REPORT TYPE AND DATES COVERED</b> Annual (Jul 1, 2002 - Jun 30, 2003)	
<b>4. TITLE AND SUBTITLE</b> New Agents for Taxol-Resistant Breast Adenocarcinoma			<b>5. FUNDING NUMBERS</b> DAMD17-00-1-0313	
<b>6. AUTHOR(S)</b> Jim Klostergaard, M.D., Ph.D.				
<b>7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)</b> The University of Texas M.D. Anderson Cancer Center Houston, Texas 77030  E-Mail: jklostergaard@mdanderson.org			<b>8. PERFORMING ORGANIZATION REPORT NUMBER</b>	
<b>9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)</b> U.S. Army Medical Research and Materiel Command Fort Detrick, Maryland 21702-5012			<b>10. SPONSORING / MONITORING AGENCY REPORT NUMBER</b>	
<b>11. SUPPLEMENTARY NOTES</b>				
<b>12a. DISTRIBUTION / AVAILABILITY STATEMENT</b> Approved for Public Release; Distribution Unlimited				<b>12b. DISTRIBUTION CODE</b>
<b>13. ABSTRACT (Maximum 200 Words)</b>  Taxol has proven to be very active in breast cancer; however, evidence for resistance to Taxol has emerged. One approach to overcoming drug resistance involves drug copolymers. The paclitaxel copolymer, PGA-TXL, has shown both reduced toxicity and greater tumor localization in animal models. It has also demonstrated reduced toxicity and greater ease of administration compared to Taxol in the clinic, and has shown activity in patients with Taxol-refractory tumors. MDA-MB-361 human breast adenocarcinoma cells were implanted orthotopically in the mammary fat pad of female nude mice. When tumor volumes reached ~20 mm <sup>3</sup> , one group was treated with PGA-TXL, 180 mg/kg i.p. Two other groups received a multiple-dose regimen of either 5 or 10 mg/kg Taxol, i.p. Control tumor volumes increased 5.89 +/- 0.43-fold (mean +/- SEM) over the next 35 days. Treatment with PGA-TXL was highly efficacious: the increase in tumor volumes in this group was 2.96 +/- 0.31-fold during the same time period (p = 0.0006). The two Taxol-treated groups failed to demonstrate significant responses: tumor volumes increased 6.06 +/- 0.25 -fold (p = 0.74) and 4.29 +/- 0.61 -fold (p = 0.101) for the 5 and 10 mg/kg groups, respectively. The key results from this study indicates that even single-dose PGA-TXL is active against MDA-MB-361, an orthotopically-implanted human Her-2/neu over-expressing breast tumor model that is highly resistant to a multiple-dose regimen of Taxol. Our pre-clinical studies suggest that among the patients who could be considered for trials with PGA-TXL are those with tumors over-expressing HER-2/neu and refractory to conventional taxanes.				
<b>14. SUBJECT TERMS</b> PGA-TXL, HER-2/neu, drug resistance, orthotopic, human breast adenocarcinoma			<b>15. NUMBER OF PAGES</b> 14	
			<b>16. PRICE CODE</b>	
<b>17. SECURITY CLASSIFICATION OF REPORT</b> Unclassified	<b>18. SECURITY CLASSIFICATION OF THIS PAGE</b> Unclassified	<b>19. SECURITY CLASSIFICATION OF ABSTRACT</b> Unclassified	<b>20. LIMITATION OF ABSTRACT</b> Unlimited	

## Table of Contents

Cover.....	1
SF 298.....	2
Table of Contents.....	3
Introduction.....	4
Body.....	5
Key Research Accomplishments.....	8
Reportable Outcomes.....	8
Conclusions.....	13
References.....	13
Appendices.....	none

## INTRODUCTION

Taxol has proven to be a valuable addition to the chemotherapeutic regimens that can be offered to breast cancer patients; however, as with other drugs, evidence for resistance to Taxol has emerged. Among these resistance mechanisms is the P-gp170 membrane-associated drug-efflux pump, for which the active agent, paclitaxel, is a substrate, and over-expression of the oncogene, HER-2/neu. Both of these resistance mechanisms are widely associated with breast cancer. Strategies to address Taxol-resistance include its combination with other chemotherapeutic agents and dose-intensification. However, in recent randomized clinical trials, the latter has proven to be largely ineffective, with little meaningful clinical benefit at the price of severe toxicities. Therefore, new agents and strategies are urgently needed to address Taxol-resistant breast cancer.

One approach to overcoming drug resistance is the use of drug copolymers. These high molecular weight conjugates can be transported via endocytosis to the endosome, where they are then cleaved to release free drug. For DNA-targeting drugs, this could afford superior nuclear access compared to import via diffusion as occurs with free drug. Further, it restricts the gradient of export of conjugate-released drug via membrane-localized drug efflux mechanisms that are clearly operant on free drug. In vivo, other considerations may be even more relevant, including distribution to tumor vs. normal tissue. High molecular weight drug copolymers may, on the one hand, 1) restrict diffusion-controlled uptake by normal tissues that occurs with free drug; but, on the other, they may 2) enhance extravasation across the abnormal tumor endothelium, thereby enhancing tumor localization compared to free drug. The paclitaxel copolymer employed in these studies, PGA-TXL, now commercially named "Xyotax", has shown both reduced toxicity and greater tumor localization in animal models, thereby fulfilling two expectations of copolymer behavior. To date, it has also demonstrated reduced toxicity and greater ease of administration compared to Taxol in the clinic, and has shown activity in patients with Taxol-refractory tumors. In this proposal, we will establish the toxicity, pharmacokinetics and anti-tumor efficacy of this Taxol copolymer in human breast adenocarcinoma models in nude mice; these models of P-gp- and HER-2/neu-mediated resistance will test the potency of the copolymer against resistance mechanisms that are operant against Taxol itself.

## BODY

### Task 1 Mechanistic Studies: Effects on Cell Cycle Distribution/Apoptosis and RAF-1 Kinase Activation

- a) Conduct cell-cycle (PI staining) and apoptosis assays (TUNEL and hypodiploidy) on human breast adenocarcinoma cell lines (P-gp and HER-2/neu models) to be used in Task 4 to establish responses to Taxol and PGA-TXL in vivo.
- b) Using these cell lines and the doses established as relevant to previous endpoints, determine role of Raf-1 kinase pathway in these responses.

These studies are currently underway with human HER-2/neu over-expressing MDA-MB-361 breast adenocarcinoma cells.

a) In initial studies with another apoptosis-inducing agent, dimethylsphingosine, we have observed that MDA-MB-361 cells rapidly, dose-dependently and progressively acquire PI-positivity, as well as positivity using the CaspaTag flow cytometric assay for pan-caspase activation. The CaspaTag-positive/PI-negative population increased by as much as ~30-fold compared to untreated controls, whereas the PI-positive population increased ~4-5-fold. Curiously, the increase in the CaspaTag-positive population appeared to initially lag behind that of the PI-positive population.

We anticipate similar patterns with regard to development of PI-staining and CaspaTag-staining following treatment with Taxol and PGA-TXL.

b) We have first undertaken these studies in a human ovarian carcinoma cell line, NMP-1, and are now conducting counterpart studies in MDA-MB-361 cells.

We have observed that the response to Taxol in NMP-1 cells is concentration- and time-dependent: 5 nM Taxol has minimal effects on cell cycle distribution at 24 hr, although it somewhat increases the hypodiploid population; this progresses to a robust G2/M block by 48 hr. At 25 nM, Taxol causes dramatic increases in the hypodiploid population as well as G2/M arrest, already by 24 hr.

The benzoquinone ansamycin, geldanamycin, which binds to the ATP binding pocket of HSP90, thereby inducing the degradation of proteins chaperoned by this HSP, has minimal effects on the cell cycle distribution of NMP-1 cells by itself, although it quenches S-phase cells at both 24 and 48 hr. When combined with 5 nM Taxol, it also appeared to quench the S-

phase population, without reducing the hypodiploid fraction induced by Taxol at 24 hr. When combined with 25 nM Taxol, geldanamycin markedly suppressed the development of hypodiploid cells induced by this concentration of Taxol, indicating the requirement for HSP90-chaperoned proteins in the apoptotic response to Taxol. Studies by Torres and Horwitz (2) in a human lung carcinoma cell line have underscored a role for Raf-1 kinase in Taxol-induced apoptosis in this model; we will pursue this aspect in MDA-MB-361 cells should their response mirror this pattern.

We have also examined the CaspaTag response of NMP-1 cells to Taxol. At 4 hr, there was a ~50% increase in the CaspaTag-positive/PI-negative population in response to 5 nM Taxol, with minimal increases in PI-positive cells observed at this time; by 20-24 hr, there was a four-fold increase in the former population, and a ~60% increase in the latter (PI-positive) population. These results are consistent with induction of the caspase cascade preceding the development of hypodiploidy. Similar studies are underway in MDA-MB-361 cells.

#### Task 2 Pharmacokinetics: Cellular and IP Administration

- a) Establish parameters of cellular uptake and fate of paclitaxel and PGA-TXL, using compounds <sup>3</sup>H-labeled in paclitaxel moiety or in PGA backbone; determine extent and site of PGA-TXL esterolysis to paclitaxel
- b) Establish pharmacokinetic parameters for peritoneal clearance of paclitaxel and PGA-TXL following i.p. administration; determine parameters for resultant plasma levels compared to i.v. administration; determine extent and site of PGA-TXL esterolysis to paclitaxel

Studies related to this task have not yet been initiated.

#### Task 3 Toxicity Studies: Single- and Multiple-Dose IP and IV MTDs

- a) Determine single-dose i.v. and i.p. MTD for PGA-TXL in nude mice

#### Task 4 Efficacy Studies: Her-2/neu- and P-gp-Mediated MDR Models HER-2/neu-mediated MDR

- a) Establish tumor responses and effects on survival of Taxol or PGA-TXL administered at single- or multiple-dose MTDs to nude mouse models of HER-2/neu high and basal expressing human breast adenocarcinomas

Initial activities relevant to these two Tasks were presented in the previous annual report; new studies pertinent to Task 4 are presented

below. The intent of the current study was to compare the anti-tumor efficacy of Taxol vs. PGA-TXL in the MDA-MB-361 model.

Poly(L-glutamic acid)-paclitaxel (PGA-TXL) was prepared by carbodiimide-mediated coupling of paclitaxel and poly(L-glutamic acid). Formulations of the final product contained ~20% paclitaxel (w/w), with a PGA backbone of ~30-40 k Da.

MDA-MB-361 human breast adenocarcinoma cells were obtained from the ATCC and were cultured exactly according to the ATCC-defined conditions and using their specific recommended serum. The cells were maintained in Liebowitz L-15 medium in the absence of CO<sub>2</sub>. This allowed retention of original cellular morphology and growth pattern in vitro, albeit with a long doubling time (~7 days). Cells were finally trypsinized and adjusted to an inoculum cell number of 4-6 X 10<sup>6</sup> viable cells.

MDA-MB-361 cells were implanted under aseptic conditions in the mammary fat pad of 5-8 week old female nude mice. When tumor volumes reached a group average of ~20 mm<sup>3</sup> (Day 28 post-implantation; see below), one group of inoculated mice was treated with PGA-TXL. The formulation was injected i.p. in 100-200 microliters volume of PBS. A single dose level of 180 mg/kg was administered one time only. Two other groups received a multiple-dose regimen of either 5 or 10 mg/kg Taxol, near the MTD, and also administered i.p. Controls were given saline. Tumor outgrowth was evaluated by caliper measurement of perpendicular tumor diameters in treated and control groups. Animals with regressing tumors, in *either* control *or* treatment groups, were excluded from further evaluation; this severe censoring eliminated *any* concern that the spontaneous regressions occasionally observed in controls would be favorably and inadvertently factored into the responses to treatment. The group-averaged volume of these tumors, calculated as length x width x width/2, was normalized to the starting volume (at time of first treatment) and plotted vs. the day of measurement, through Day 63 post-implantation (35 days post-treatment initiation).

Control tumor volumes increased 5.89 +/- 0.43 -fold (mean +/- SEM) over this 35 day interval. Treatment with PGA-TXL was highly efficacious: the increase in tumor volumes in this group (11 mice) was only 2.96 +/- 0.31 -fold during the same time period (p = 0.0006 compared to controls; unpaired t-test). In contrast, the two Taxol-treated groups failed to demonstrate significant responses: tumor volumes increased 6.06 +/- 0.25 -fold (p = 0.74) and 4.29 +/- 0.61 -fold (p = 0.101) for the 5 and 10 mg/kg groups, respectively. However, their responses differed from each

other ( $p = 0.036$ ) and the higher dose group nominally failed to be distinguishable ( $p = 0.056$ ) from the PGA-TXL group. Therefore, and in the absence of evidence for toxicity with the 10 mg/kg dose level, in a future experiment to be conducted during the un-funded extension year, we will assess the response of the 361 model to a higher Taxol dose, likely 15 mg/kg.

### KEY RESEARCH ACCOMPLISHMENTS

- The key results from this study indicate that even single-dose PGA-TXL is active against MDA-MB-361, an orthotopically-implanted human Her-2/neu over-expressing breast tumor model. Of note, this model appeared highly resistant to a multiple-dose regimen of Taxol.

### REPORTABLE OUTCOMES

Three abstracts/presentations have arisen from this work since the commencement of the grant, and following verification and extension to a higher dose level of Taxol in the MDA-MB-361 model, we will prepare a manuscript.

1) The following abstract was submitted and accepted to the 6<sup>th</sup> US-Japan Symposium on Drug Delivery Systems, held in December, 2001 in Maui, HI. It was presented as a poster as well as being selected for presentation in a workshop.

### PACLITAXEL COPOLYMER TO ADDRESS TAXOL RESISTANCE

J. Klostergaard, E. Auzenne, C. Li, N.J. Donato, M. Khodadadian, D. Farquhar, and Y. Zou

The University of Texas M.D. Anderson Cancer Center  
Houston, Texas 77030 USA

We have evaluated a paclitaxel-poly(L-Glu) copolymer in human tumor/nude mouse orthotopic xenograft models which either reflect resistance to Taxol (HEY/ovarian) or over-express HER-2/neu (MDA-361/breast). Early treatment (Day 2 HEY) with MTD Taxol achieved some improvement in survival, but was not curative. However, treatment with copolymer markedly improved survival and some apparent cures were observed. The higher tumor burden at Day 7 rendered this model resistant to MTD Taxol, but still responsive to copolymer. Similarly, early treatment (Day 7) of the 361 breast model with paclitaxel copolymer resulted in substantial tumor growth delay, regression, or even apparent cure. When administered later, the copolymer still caused tumor growth delay, but no cures were observed. We conclude that

formulation of paclitaxel with this poly(L-Glu) backbone substantially enhanced its potency, and rendered it active in two highly drug-resistant models. Supported in part by DOD grants BC980420, BC991113 and OC000036 (JK).

2) The following abstract was presented as a poster at the Era of Hope Meeting, sponsored by the DOD Breast Cancer Research Program, in Orlando, FL, September 25th-28th, 2002.

LIPOSOMAL-DIMETHYL-SPHINGOSINE AND PACLITAXEL COPOLYMER ARE ACTIVE AGAINST HER-2/NEU OVER-EXPRESSING HUMAN BREAST ADENOCARCINOMA ORTHOTOPIC XENOGRAFT MODEL

Jim Klostergaard, Ph.D., Edmond Auzenne, M.S., C. Li, Ph.D., M. Khodadadian, B.S., David Farquhar, Ph.D. and Yiyu Zou, Ph.D.

The University of Texas  
M.D. Anderson Cancer Center  
Houston, TX 77339

E-mail: jkloster@mdanderson.org

Over-expression of HER-2/neu has been linked to poorer prognosis and survival in breast cancer patients. The basis for this association likely includes therapeutic resistance, including resistance to Taxol (paclitaxel), widely used in many chemotherapeutic regimens for this disease. We have recently observed that certain sphingolipids, either as free lipids or as constituents of liposomes, induce apoptosis *in vitro* in tumor cells despite the over-expression of Her-2/neu. Further, we have reported that a paclitaxel copolymer, paclitaxel-poly(L-glutamic acid) (PGA-TXL), is active against Taxol-resistant tumors *in vivo*.

We therefore evaluated liposomal-dimethyl-sphingosine (L-DMSP) and PGA-TXL in a human HER-2/neu over-expressing breast adenocarcinoma (MDA-361) orthotopic xenograft model. Tumor cells ( $4-6 \times 10^6$ ) were implanted in the mammary fat pad of 5-8 week old female nude mice. Mice were treated *i.p.* either one-week later or when tumors grew to 5-6 mm diameter.

Early treatment with a multiple-dose regimen of L-DMSP (4.5 mg DMSP per dose; 20 mole percent of a small unilamellar vesicle formulation), caused a delay in or reduced subsequent tumor growth, but was not curative. However, early treatment with a single-dose of PGA-TXL (180 mg/kg paclitaxel equivalents), also one week after tumor implantation, resulted in substantial tumor growth delay, regression, or even apparent cure in two of four mice (control tumor areas at 10 weeks post-implant =  $44 \pm 21.2 \text{ mm}^2$ ; treated group areas =  $6 \pm 6.0 \text{ mm}^2$ ). When administered at the later timepoint to another group of animals, PGA-TXL still caused tumor

growth delay, but no cures were observed (treated group areas =  $24 \pm 15.3 \text{ mm}^2$ ); nor did administration of L-DMSp at this time appear to be efficacious.

We conclude that DMSP as a liposomal formulation has some efficacy against this HER-2/neu over-expressing model when the tumor burden is low. Formulation of paclitaxel with the poly(L-glutamic acid) backbone substantially reduced its toxicity, enhanced its potency, and rendered it active against this HER-2/neu over-expressing breast adenocarcinoma model.

Supported by U.S. Army Medical Research and Material Command under DAMD17-99-1-9265 and DAMD17-00-1-0313.

3) The following abstract, submitted online, was accepted for presentation as a poster at the AACR/EORTC Meeting to be held in Frankfurt, Germany on November 19-23, 2002.

Therapeutic resistance to Taxol is a major issue in a number of cancers, particularly breast and ovarian carcinoma. This resistance is multifactorial, including P-gp170-linked MDR and over-expression of HER-2/neu. We evaluated the efficacy of a paclitaxel-poly(L-Glu) copolymer (PGA-TXL) in a human ovarian carcinoma orthotopic xenograft model which reflects resistance to Taxol (HEY); we also evaluated PGA-TXL as well as a liposomal (SUV) formulation of dimethyl-sphingosine (L-DMSP; which induces apoptosis in a broad spectrum of tumor cell lines *in vitro*) in an orthotopic human breast adenocarcinoma model that over-expresses HER-2/neu (MDA-361). In the ovarian model, early treatment (Day 2 post-implantation) with multiple-dose MTD Taxol (10 mg/kg) i.p. achieved slight improvement in survival, but was not curative. However, treatment with a single dose (180 mg/kg, paclitaxel equivalents) of PGA-TXL i.p. markedly improved survival and induced some apparent cures. The higher tumor burden present on Day 7 rendered this model resistant to MTD Taxol administration at this time, but still responsive to PGA-TXL. For the breast model, treatment on Day 7, before tumors were palpable, with PGA-TXL resulted in subsequent tumor growth delay, regression, or even apparent cure. Treatment at this time with a multiple-dose regimen of L-DMSP (4.5 mg DMSP/dose) i.p., caused a delay in or reduced subsequent tumor growth, but was not curative. When administered later after tumors grew to 5-6 mm diameter, PGA-TXL still caused tumor growth delay, but no cures were observed; administration of L-DMSP at this later time was not efficacious. We conclude that formulation of paclitaxel with this poly(L-Glu) backbone substantially enhanced its potency, and rendered it active in drug-resistant ovarian and breast models. Further, we conclude that DMSP as a liposomal formulation has some efficacy against this HER-2/neu over-expressing breast model: however, only when the tumor burden is low. (Supported in part by DOD grants BC980420, BC991113 and OC000036 to JK).

## CONCLUSIONS

HER-2/neu over-expression in breast cancer portends an aggressive clinical course and greater resistance to certain therapeutic regimens, including those involving taxanes. Although the advent of Herceptin has brought new opportunities for more effective and targeted therapy for women with this marker, other approaches must also be exploited. The use of a drug copolymer strategy for paclitaxel (Taxol) based on a poly(L-glutamic acid; PGA) backbone has proven in pre-clinical and clinical studies to reduce the toxicity of paclitaxel. Importantly, activity of PGA-TXL in Taxol-resistance settings has been observed, as well.

The key inference: our pre-clinical studies suggest that among the patients who could be considered for trials with PGA-TXL are those with tumors over-expressing HER-2/neu and refractory to conventional taxanes.

## REFERENCES

1. Rowinsky EF and Donehower RC. Paclitaxel (Taxol) *New Eng J Med* 332; 1004-1014, 1995.
2. Torres K and Horwitz SB. Mechanisms of Taxol-induced cell death are concentration dependent. *Cancer Res* 58; 3620-3626, 1998.
3. Giannakakou P, Sackett DL, Kang Y-K, Zhirong Z, Buters JTM, Fojo T and Poruchynsky S. Paclitaxel resistant human ovarian cancer cells have mutant  $\beta$ -tubulins that exhibit impaired paclitaxel-driven polymerization. *J Biol Chem* 272; 17118-17125, 1997.
4. Yu D, Liu B, Tan M, Li J, Wang SS, Hung M-C. Overexpression of c-erbB2/neu in breast cancer cells confers increased resistance to Taxol via mdr-1-independent mechanisms. *Oncogene* 13; 1359-65, 1996.
5. Yu D, Liu B, Jing T, Sun D, Price JE, Singletary SE, Ibrahim N, Hortobagyi GN and Hung M-C. Overexpression of both p185c-erbB2 and p170mdr-1 renders breast cancer cells highly resistant to taxol. *Oncogene* 16; 2087-2094, 1998.
6. Rowinsky EK. On pushing the outer edge of the outer edge of paclitaxel's dosing envelope. *Clin Cancer Res* 5; 481-486, 1999.
7. Nabholz J-M, Nabholz J-M, Gelmon K, Bontenbal M, Spielmann M, Catiemel G, Conte P, Klaassen U, Namer M, Bonnetterre J, Fumoleau P and Winograd B. Multicenter, randomized comparative study of two doses of paclitaxel in patients with metastatic breast cancer. *J Clin Oncol* 14; 1858-1867, 1996.

8. Winer E, Berry D, Duggan D, Henderson CI, Cirrincione C, Cooper R and Norton L. Failure of higher dose paclitaxel to improve outcome in patients with metastatic breast cancer: results from CALGB 9342. *Proc Am Soc Clin Oncol* 17; 101a, 1997.
9. Sparreboom A, van Tellingen O, Nooijen WJ and Beijnen JH. Nonlinear pharmacokinetics of paclitaxel in mice results from the pharmaceutical vehicle Cremophor EL. *Cancer Res* 56; 2112-2115, 1996.
10. Li C, Yu D-F, Newman RA, Cabral F, Stephens C, Hunter N, Milas L and Wallace S. Complete regression of well-established tumors using a novel water-soluble poly (L-glutamic acid)-paclitaxel conjugate. *Cancer Res* 58; 2404-2409, 1998.
11. Nicoletti MI, Lucchini V, Massazza G, Abbott BJ, D'Incalci M & Giavazzi R. Antitumor activity of taxol (NSC-125973) in human ovarian carcinomas growing in the peritoneal cavity of nude mice. *Ann Oncol* 4: 151-155, 1993.
12. Eiseman JL, eddington ND, Leslie L, McAuley C, Sentz DL, Zuhowsky M, Kujiwa JM, Young D and Egorin MJ. Plasma pharmacokinetics and tissues distribution of paclitaxel in CD2F1 mice. *Cancer Chemother Pharmacol* 34; 465-471, 1994.
13. McCloskey DE, Kaufmann SH, Prestigiacomo LJ and Davidson NE. Paclitaxel induces programmed cell death in MDA-MB-468 human breast cancer cells. *Clin Cancer Res* 2; 847-854, 1996.
14. Markman M, Rowinsky E, Hakes T, Reichman B, Jones W, Lewis JL, Jr, Rubin S, curtin J, Barakat R, Hurowitz L, Almadrones L and Hoskins W. Phase I trial of intraperitoneal Taxol: a gynecologic oncology group study. *J Clin Oncol* 10; 1485, 1992.