ADXS11-001 immunotherapy targeting HPV-E7: Final results from a Phase 2 study in Indian women with recurrent cervical cancer

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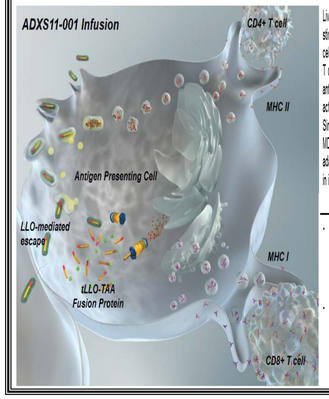
Abstract

Background: ADXS11-001 immunotherapy is a live attenuated *Listeria monocytogenes* (*Lm*) bioengineered to secrete a HPV-16-E7 fusion protein targeting HPV transformed cells. The *Lm* vector serves as its own adjuvant and infects APC where it cross presents HPV-E7-tLLO fusion peptide, stimulating MHC class 1 and 2 pathways resulting in specific T-cell immunity to tumors. Here we describe audited final results from *Lm*-LLO-E7-015, a randomized P2 study designed to evaluate the safety and efficacy of ADXS11-001 with and without cisplatin in 110 patients with recurrent cervical cancer in India; who had recurred after prior cytotoxic therapy, including chemotherapy, radiotherapy or both. **Methods:** Patients were randomized to either 3 doses of ADXS11-001 at 1 x 10^9 cfu or 4 doses of ADXS11-001 at 1 x 10^9 cfu with cisplatin chemotherapy (40 mg/m^2). Naprosyn and oral promethazine were given as premedications and a course of ampicillin was given 72h after infusion to clear the vector. Patients received CT scans at baseline and 3, 6, 9, 12 and 18 months. The primary endpoint was overall survival. **Results:** The final audited irRECIST 12 month survival was 32% (35/109) and 18-month survival was 22% (24/109). The response rate was 11% (5 CRs and 6 PRs/110) with tumor responses observed in both treatment arms; 31 additional patients had stable disease ≥ 3 months, for a disease control rate of 38% (42/110). Average duration of response in both ADXS11-001 containing treatment groups was 9.5 months. Long-term survival >24 months was observed in 18% (16/91) patients. Activity against different high-risk HPV strains was observed. The incidence of SAEs possibly related or related to ADXS11-001 was 1% G3 (0% G4-5). The majority of non-serious adverse events were predominately infusion associated, and either resolved on their own or responded to symptomatic treatment. **Conclusions:** ADXS11-001 appears to have significant clinical activity in patients with recurrent cervical cancer. Clinical benefit includes prolonged survival and objective tumor responses, including complete responses. The addition of cisplatin to ADXS11-001 did not significantly improve survival outcomes or tumor responses. These results were observed with 1 cycle of ADXS11-001 at the lowest effective dose. The baseline ECOG performance status, type of prior therapy, or aggressiveness of disease had no effect on survival outcomes and tumor responses. The 32% 12 month survival, 22% 18 month survival, and 11% response rate observed in this recurrent disease setting compares favorably with more toxic chemotherapy treatment options. Further clinical development includes optimization of the ADXS11-001 dose and schedule including multiple cycles of treatment, use in combination, and sequencing with other agents. Based on these data, this agent is being considered for future registration studies.

Lm-LLO Immunotherapy

- ADXS11-001 is a live attenuated bioengineered *Listeria monocytogenes* (*Lm*) LLO immunotherapy for the treatment of HPV-associated cancer
- ADXS11-001 secretes an antigen-adjuvant fusion protein consisting of a truncated fragment of the *Lm* listeriolysin (tLLO) fused to HPV16-E7
- *Lm*-LLO immunotherapy redirects the potent inherent cellular immune responses to *Lm* toward cells expressing the tumor associated antigen (TAA)
- *Lm*-LLO immunotherapy provides a comprehensive system for generating a cellular immune response:
  - Powerful innate immunity: TLRs, NOD-1, 2, PAMP; no adjuvant required
  - Access to APC: Cross presents tumor antigen
  - Powerful Adaptive immunity: Antigen specific CD4+, CD8+ T cells
  - Reduction of immunologic tolerance (Tregs and MDSCs) in the tumor microenvironment
  - Vector can be cleared with antibiotics

Life Cycle of *Lm* in APC

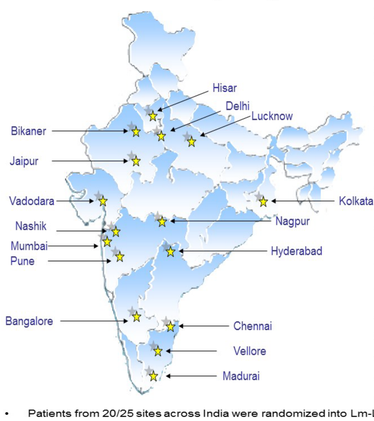


Live attenuated *Lm* bioengineered to secrete an antigen-adjuvant fusion protein (antigen + tLLO) stimulate a profound innate immune response and are selectively phagocytosed by antigen presenting cells (APC). Fragments from *Lm* are processed via the MHC class I generating antigen specific CD4+ T cells. Some *Lm* secrete LLO which enables them to escape into the cytosol where they secrete antigen-LLO fusion proteins. Fusion protein antigens are presented via MHC class I to generate activated CD8+ T cells. The activated T cells find, infiltrate tumors and destroy the tumor cells. Simultaneously, immunologic tolerance in the tumor microenvironment mediated by Treg cells and MDSCs is reduced enabling better tumor cell destruction. Thus *Lm*-LLO agents stimulate innate and adaptive tumor-specific immunity while simultaneously reducing immune tolerance to tumors resulting in improved survival and tumor responses.

Live Attenuated *Listeria monocytogenes*

- Attenuation: Genetically Engineered
  - Loss of bacterial virulence due to 10,000 to 100,000 fold attenuation
    - Deletion of p.prfA (with D133V complementation) results in reduction of bacterial virulence factors
    - Recombination and restoration of virulence not possible
  - Secretes HPV-E7 protein fused with highly immunogenic tLLO fragment within cytoplasm of APC leading to antigen-specific T-cell immunity
- *Lm*-LLO agents are nonpathogenic, consistent with **BSL-1 and RG1 agents**
  - According to:
    - US Centers for Disease Control (import and shipping permits)
    - German ZKBS (manufacturing)
  - Published data has shown that there is no difference in the kinetics of clearance in wild-type or IFN-γ knockout mice
  - SCID mice clear Lm-LLO agents at doses 100,000x the LD50 of wild type Lm in normal mice.

Participating Sites in India



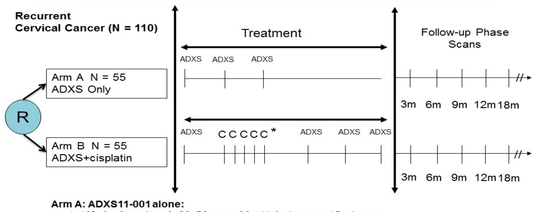
S. No.	Site	PI
100	Ruby Hall Clinic, Pune	Dr. Minish Jain
101	Curie Manavata Cancer Centre, Nashik	Dr. Rajnish V. Nagarkar
103	Central India Cancer Research Institute, Nagpur	Dr. Ajay Mehta
104	M V Hospital & Research Centre, Lucknow	Dr. Vijay Kumar
105	Dr. Kamakshi Memorial Hospital, Chennai	Dr. Sumana Premkumar
107	Tata Memorial Centre, Parel	Dr. Sudeep Gupta
109	Bibi General Hospital & Cancer Center, Hyderbad	Dr. Sudha Sinha
110	Chittaranjan National Cancer Institute, Kolkata	Dr. Partha Basu
111	Dr. Ida B Scudder Cancer Centre, Vellore	Dr. Subhashini John
112	Deenanath Mangeshkar Hospital & Research Centre,Pune	Dr. Chetan Deshmukh
113	King George Medical University, Lucknow	Dr. Kirti Srivastava
115	Meenakshi Mission Hospital and Research Centre, Madurai	Dr. Kirushna Kumar
116	Nak Hospital Baroda	Dr. Vibha Nak
117	S. K. Soni Hospital, SEAROC Cancer Centre, Jaipur	Dr. Aniah Maru
118	Indo American Cancer Institute and research centre, Hyderabad	Dr. Senthil Rajappa
119	Smt. Kashibai Navale Medical College and General Hostal Pune	Dr. Rakesh Neve
121	SMS Medical College and Hospital, Rajasthan	Dr. Om Prakash Sharma
124	Acharya Tulsi Regional Cancer Treatment and Research Institute	Dr. Ajay Sharma
126	Narayana Hrudayalaya Hospital	Dr. Mridul Gehot
128	Nanja Gutt Memorial Cancer Hospital,	Dr. Prakash Dale

- Patients from 20/25 sites across India were randomized into Lm-LLO-E7-15
- Advaxis extends their gratitude and acknowledgement to the investigators, the site teams, and especially the patients for their participation in Lm-LLO-E7-15

Lm-LLO-E7-015: A Randomized Phase 2 Study to Assess the Safety & Efficacy of ADXS-HPV +/- cisplatin Treatment for Recurrent Cervical Cancer

- **20 sites throughout India**
- **N=110**:
  - Women 18-60 years of age with recurrent or refractory cervical cancer who have recurred after prior therapy (radiation therapy +/- chemotherapy)
  - ECOG performance status 0-2
  - Randomized - 2 groups of 55 patients receiving: ADXS11-001 or ADXS11-001 + cisplatin
- **Primary Objective:**
  - To determine the safety and efficacy ADXS11-001 +/- cisplatin
- **Efficacy Endpoints:**
  - Primary efficacy endpoint is overall survival.
  - Secondary efficacy endpoints are tumor response (RECIST 1.1) and PFS
- **Immunologic Evaluations:**
  - Serum cytokines, HPV specific T cells, and PBMC phenotyping

Trial Design: Lm-LLO-E7-15



Arm A: ADXS11-001 alone:
- 1x10^9 cfu x3 on days 0, 28, 56 as an 80 ml infusion over 15 minutes

Arm B: ADXS-HPV + cisplatin:
- ADXS11-001 = 1x10^9 CFU as an 80 ml infusion over 15 minutes on days 0, 88, 106, 134
- *cisplatin = 40 mg/m^2 x5 weekly on days 30, 37, 44, 51, 58

Lm-LLO-E7-015 was designed to evaluate the safety and efficacy of ADXS11-001 given as monotherapy or with cisplatin. The ADXS11-001 treatment arm received ADXS11-001 (1x10^9 cfu) as 3 IV infusions 4 weeks apart, each dose followed by antibiotic at 3 days post-dosing. The ADVX11-001 + cisplatin treatment arm received ADXS11-001 as an IV infusion (1x10^9 cfu), followed by antibiotic beginning 3 days post-dosing, followed 4 weeks later with 5 weekly IV administrations of cisplatin (40 mg/m^2) followed 4 weeks later by 3 IV infusions of ADXS11-001 one month apart with antibiotic beginning 3 days after each ADXS11-001 dose. Naproxsyn 500 mg BID, (Day -1, 0) and promethazine 25 mg PO, BID (pre-dose, 8 hours) were administered as premedications. Ampicillin 500 mg QID (Days 3-9) is administered post-infusion. Safety was assessed at every visit. Efficacy was determined from overall survival and scans taken at baseline (before the first treatment dose) and at 3, 6, 9 12, & 18 months after treatment. Patients were are followed for survival for duration of the study.

General Demographics

	Overall	ADXS-HPV (n=55)	ADXS-HPV + Cisplatin (n=55)
Aggressive (Recurred <24M)	80%		
Squamous	100%		
Prior Platinum Chemotherapy	58% (63/109)		
Prior Chemo for Recurrence (2nd Line)	17% (19/109)		
Stage IV	24% (26/109)	24% 13 (7A/6B)	24% 13 (4A/9B)
Stage III	39% (43/109)	35% 19 (5A/14B)	44% 24 (7A/17B)
Stage II	22%(24/109)	25% 14 (5A/9B)	18% 10(4A/6B)
Stage IB	14% (15/109)	13% 7	15% 8
Primary Therapy	CT/RT: 37% (40/109) CT: 11% (12/109) RT: 52% (57/109)	CT/RT: 36% (20/55) CT: 0% (0/55) RT: 65% (36/55)	CT/RT: 33% (18/54) CT: 20% (11/54) RT: 44% (24/54)
ECOG Status *(all randomized patients who received at least one dose n=109)*	0: 40% (35/109) 1: 49% (64/109) 2: 11% (12/109)	0: 40% (22/55) 1: 49% (29/55) 2: 11% (6/55)	0: 24% (13/54) 1: 65% (35/54) 2: 11% (6/54)

HPV Strains: HPV16 =70%, HPV18 = 16%, HPV33, 35, 6 = 2% each, HPV45 =1

- **110 patients were randomized and 109 patients received at least 1 dose of ADXS11-001**
- **The majority of patients had a poor prognosis:**
  - 60% were ECOG status 1-2 at baseline
  - 63% were Stage 3 or 4 at initial diagnosis
  - 80% Aggressive disease (Recurred< 24 M)
  - 87% had prior pelvic EBRT
  - 58% Prior platinum chemotherapy
  - 17% Failed prior chemotherapy for recurrent cervical cancer (2nd Line)

Safety Summaries: Lm-LLO-E7-15

109 patients received 264 doses of ADXS11-001 at 1x10^9 cfu

AEs *related or possibly related* to study drug:

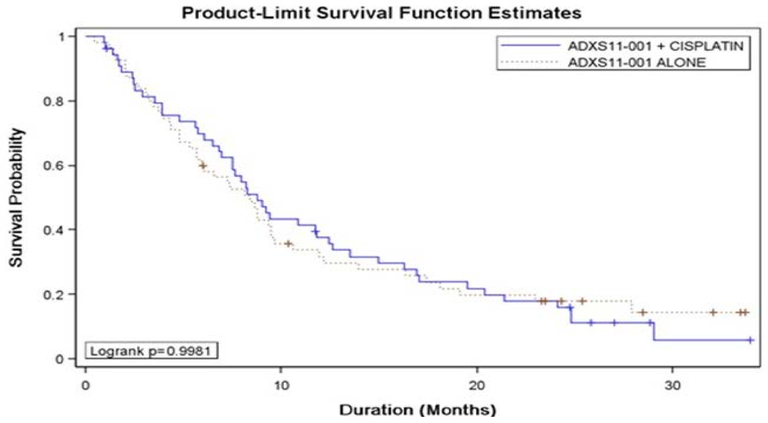
- **41 patients (38%) reported 76 Grade 1-2 AEs**
  - 41 Chills/Shivering
  - 13 Flu Like Symptoms
  - 6 Vomiting
  - 5 Nausea
  - 5 Fever
  - 2 Dizziness
  - 1 Cytokine Release Syndrome
  - 1 Headache
  - 1 Weight Decreased
  - 1 Blood Alkaline Phosphatase Increased
- **1 Grade 3 AE reported as Fever**
- **0 Grade 4 AEs**
- **0 Grade 5 AEs**

ALL AEs (Related and Unrelated to ADXS11-001)

- **95 patients (87%) experienced 653 AEs**
- **49 patients (45%) experienced at least one SAE (67/653)**
  - 21 Disease Progression
  - 8 Anemia
  - 9 Renal Failure (4 Obstructive Uropathy)
  - 5 Death (Sudden/Unknown Cause)
  - 5 Haemorrhage
  - 3 GI Obstruction
  - 1 SAE each: Abdominal Pain, Athralgia, Cardiopulmanary Failure, Cytokine Release Syndrome, Deep Vein Thrombosis, Dyspnoea, Pyrexia, Gastritis, Hypothermia, Intestinal Perforation, Multi-Organ Failure, Peritonitis Bacterial*, Pulmonary Embolism, Psychotic Disorder, Renal Injury, Urinary Tract Infection, Vomiting

**E. coli*

KM Curve & Overall Survival



		Treatment Group	
Patients at Risk, n (%)*	Overall (N=109)	ADXS11-001 ALONE (N=55)	ADXS11-001 + CISPLATIN (N=54)
12 Months			
n	109	55	54
% alive (#)	32% (35)	29% (16)	35 % (19)
18 Months			
n	109	55	54
% alive (#)	22% (24)	22% (12)	22% (12)
24 Months (Preliminary)			
n	96	44	42
% alive (#)	18% (16/91)	16% (7/44)	21% (9/42)

The primary endpoint of OS is calculated from the efficacy population-survival-follow-up of 109 and is defined as all patients who had been treated with at least one dose of ADXS11-001

The Kaplan Meier curve above represents overall survival for all patients.

- The addition of cisplatin to ADXS11-001 did not significantly improve survival (p=0.9981)
- Median overall survival was 8.6 months but mean 9.5 months, suggesting a subgroup of long-term survivors
- No differences in overall survival were observed based on:
  - Prior therapy of combination chemotherapy and radiation, radiation alone, or chemotherapy alone
  - Aggressiveness of disease
  - ECOG Status at baseline

- 12 month overall survival of 32% (35/109) and 18 month survival of 22% (24/109) are notable in this disease setting and are consistent with an active agent in recurrent cervical cancer.
- Preliminary 24 month overall survival of 18% (16/91) suggests a subgroup of patients who experienced long-term survival.

CR and PR Case Studies
CT Scan Evaluation at 3, 6, 9, 12 and 18 months

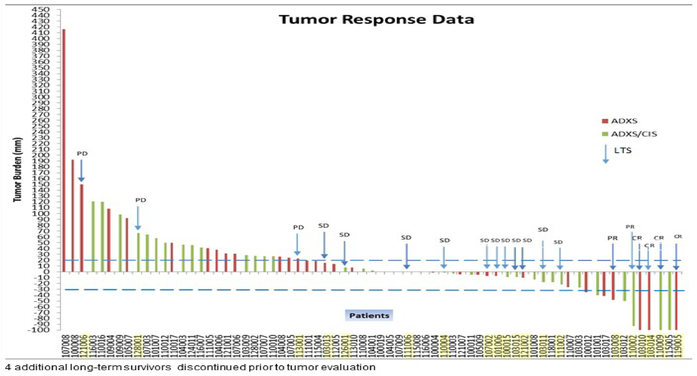
All patients who achieved a CR/PR in Lm-LLO_E7-15:

Patient #	First Line Tx	Stage	Tx Arm	Tumor Burden (mm)						Tumor Decrease
				Baseline	3 mo.	6 mo.	9 mo.	12 mo.	18 mo.	
Complete Responses										
119-005	RT	IIIA	ADXS	37	35	0	30	26	0	100%
115-005	RT	IIB	ADXS + CIS	30	0	0	0	0	EXP. 15 mo.	100%
110-009	CT + RT	IB	ADXS + CIS	23	0	0	0	0	0	100%
103-014	CT	IVB	ADXS	223	228	0	DC 10/22/12	-	-	100%
103-010	CT	IVA	ADXS	35	0	0	0	0	DP 15 mo.	100%
Partial Responses										
110-002	RT	IVB	ADXS + CIS	284	84	56	34	20	36	93%
101-001	CT + RT	IVB	ADXS + CIS	50	42	44	20	EXP. 11.5 mo.	-	60%
103-012	CT + RT	IVB	ADXS + CIS	18	9	25	WC 10/22/12	-	-	50%
103-008	RT	IVA	ADXS	48	48	39	39	25	28	48%
103-017	CT + RT	IVB	ADXS	106	62	EXP 3.1 mo.	-	-	-	41%
100-012	CT + RT	IVB	ADXS	164	107	105	105	EXP. 9.5 mo.	-	36%

DP = Disease Progression; EXP = expired; NA = Not Available; WC = Withdrew Consent

- 5 complete responses and 6 partial responses have been observed in 69 evaluable patients
- ADXS11-001 alone resulted in 2CR and 3PR. ADXS11-001 + cisplatin resulted in 3 CRs and 3 PRs
- Objective responses were seen in patients with bulky disease and multiple metastases
- Responses were seen in patients previously treated with combination chemotherapy/radiotherapy, radiotherapy alone or chemotherapy alone

Lm-LLO-E7-15 Tumor Response Data



4 additional long-term survivors discontinued prior to tumor evaluation

The waterfall plot above depicts the best overall response for patients evaluable at ≥3 months (69/110). 41 patients discontinued prior to the first tumor evaluation (16 withdrew consent, 15 expired, 5 were lost to follow up, 4 discontinued, and 1 excluded for inconsistent radiography)

- Objective CR's, PR, numerous minor responses and stable disease ≥3 months observed
- Disease Control Rate (CR + PR + SD) = 38% (42/109)
- Using irRECIST criteria 11 patients had objective responses (5CR/6PR), 31 patients had stable disease ≥ 3 months, 27 patients had progressive disease.
- The disease control rate was 38% (42/1109)
- The addition of cisplatin chemotherapy did not improve tumor responses.
- Tumor responses were observed in patients infected with different high risk HPV strains including HPV16, 18, 31, 33 and 45

Long-Term Survivors (LTS)

- 22% (24/109) of patients are long-term survivors (alive >18 months, range 18-34 months and are indicated by the blue arrows)
- 4 long term survivors discontinued prior to tumor evaluation

Patient Demographics of Long Term Survivors (≥18M)

Patient #	First Line Tx	# Prior Tx	Stage	ECOG	Tx Arm	BOR	# Months Alive
103-008	RT	1	IVA	0	ADXS	-48%	36.07
103-010	CT	1	IVA	1	ADXS	-100%	34.68
115-007	RT	1	IIB	0	ADXS + CIS	WC	34.00
113-001	RT	1	IIIB	1	ADXS	WC	33.70
110-009	RT	1	IB	1	ADXS + CIS	-100%	31.46
103-013	RT(1), CT(2)	2	IVB	0	ADXS	WC	31.17
103-015	CT	1	IVB	1	ADXS + CIS	-9%	29.72
111-002	RT	1	IIB	1	ADXS + CIS	-22%	28.67
103-014	CT	1	IVB	0	ADXS	-100%	27.55
126-001	CT/RT	1	IIIA	2	ADXS + CIS	+7%	27.42
119-003	RT	1	IB	1	ADXS + CIS	WC	25.45
119-005	RT	1	IIIA	1	ADXS	-100%	25.05
100-015	CT/RT	1	IIIB	0	ADXS + CIS	-9%	24.46
124-005	RT	1	IB	2	ADXS + CIS	WC	24.46
121-002	CT/RT	1	IB	0	ADXS	-10%	24.00
103-011	CT/RT	1	IVB	0	ADXS + CIS	-20%	23.80
111-006	RT	1	IVA	1	ADXS	-100%	23.50
121-006	CT/RT	1	IB	0	ADXS	+50%	22.98
126-002	CT/RT	1	IIIB	2	ADXS	WC	22.65
128-001	RT(1), CT(2)	2	IIA	0	ADXS + CIS	+60%	21.14
110-002	RT	1	IVB	1	ADXS + CIS	-93%	20.09
110-004	RT	1	IIIB	1	ADXS + CIS	-1%	19.23
101-006	CT/RT	1	IIB	0	ADXS	-21%	18.87
107-002	CT/RT	1	IIB	1	ADXS	-7%	18.10

Long-term survivors in recurrent cervical cancer are rare. ADXS11-001 is the first immunotherapy in cervical cancer to be associated with objective tumor responses (including CR's and PR's) and with long term survival as a monotherapy or in combination with cisplatin.

The unique mechanism of action of this immunotherapy leads to some interesting observations in the demographics of the long term survivors.

Long-term survival was observed in 22% (24/109) of patients who were treated with a well-tolerated immunotherapy associated with minimal infusion-related side effects.

This survival was observed after treatment with 1 cycle (3 doses) of ADXS11-001 given at the lowest effective dose (1x10^9 CFU):

- 25% (3/12) of patients with ECOG 2 baseline experienced long term survival
- 8% of the long term survivors (2/24) received ADXS11-001 as second line treatment for recurrent cervical cancer
- LTS was associated with some degree of tumor reduction in 14/24 patients but 6 patients showed tumor increases. 4 other withdrew consent or discontinued prior to the first evaluation
- LTS were evenly distributed between both treatment groups
- 18% (16/91) patients were alive >24 months (range 24-34+ months)

Conclusions

- **Safety**
  - ADXS11-001 was well tolerated with mild transient adverse events observed in 38% (41/109) of patients associated with infusion. All observed adverse events either self-resolved or responded readily to symptomatic treatment.
  - 1 Grade 3 SAE observed in 254 doses administered to 109 patients
- **Survival**
  - 32% (35/109) of patients were alive at 12 months; 22% (24/109) of patients were alive at 18 months
  - 18% of patients (16/91) survived >24 months (range 24-34+months)
  - Addition of cisplatin chemotherapy did not significantly improve survival or tumor response
  - Treatments received prior to entering the trial had no impact on survival or tumor response
  - Aggressiveness of disease had no impact on survival or tumor response
  - 58% of long term survivors had a baseline ECOG performance status of 1 (46%) or 3 (13%)
- **Tumor Responses are Equivalent in Both Treatment Groups**
  - 11% objective response rate (including CRs and PRs), disease control rate of 38% (42/109)
  - Combination with cisplatin did not improve the response rate
  - Median duration of response 9.5 months
  - Tumor response was not affected by prior therapy, aggressiveness of disease or ECOG status at baseline
- **Activity in Patients with Various Different High Risk HPV Strains**
  - Tumor responses observed in patients infected with all high risk HPV strains detected, including HPV16, 18, 31, 33, and 45
- **Long-term survival 18% >24 months, 18 month survival of 22%, and an 12 month survival of 32% is remarkable in patients with recurrent cervical cancer and compares favorably with other active agents/regimens in this disease setting**
- **Further clinical development includes optimization of the ADXS11-001 dose and schedule including higher doses, multiple cycles of treatment, use in combination, and sequencing with other agents**
- **The potential of ADXS11-011 to improve survival in recurrent cervical cancer versus standard of care will be evaluated in an upcoming Phase 3 clinical trial**

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Lm-LLO-E7-015 is sponsored by Advaxis, Inc., Princeton, NJ
Editorial and writing support provided by Advaxis, Inc., Princeton, NJ