ADXS11-001 *Lm*-LLO Cancer Immunotherapy: Final Results and Long-Term Survival Data From a Randomized Phase 2 Study in Recurrent Cervical Cancer

Partha Basu[1], Ajay Mehta[2], Minish Jain[3], Sudeep Gupta[4], Rajnish Nagarkar[5], Vijay Kumar[6], Sumana Premkumar[7], Rakesh Neve[8], Subhashini John[9], and Robert Petit[10]

[1]Chittaranjan National Cancer Institute, Kolkata, India, [2]Central India Cancer Research Institute, [3]Ruby Hall Clinic, [4]Tata Memorial Hospital, [5]Curie Manavata Cancer Center, India, [6]MV Hospital and Research Center, [7] Dr. Kamakshi Memorial Hospital, [8] Smt. Kashibai Navale Medical College and General Hospital, [9]Christian Medical College Vellore, [10]Advaxis, Inc. Princeton, NJ

ADVAXIS IMMUNOTHERAPIES™

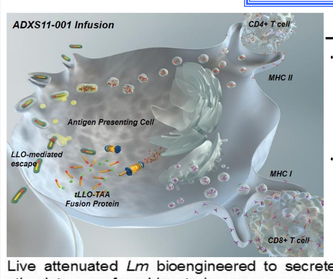
Abstract

Background: ADXS11-001 immunotherapy is a live attenuated *Listeria monocytogenes* (*Lm*) bioengineered to secrete a tLLO-HPV-16-E7 fusion protein targeting HPV-transformed cells. The *Lm* vector is its own adjuvant and infects APC where it cross presents HPV-E7-tLLO fusion protein, stimulating MHC class 1 and 2 pathways resulting in HPV-E7 specific T-cell immunity. *Lm*-LLO-E7-015 was a randomized P2 study designed to evaluate the safety and efficacy of ADXS11-001 +/- cisplatin in patients with recurrent cervical cancer in India. **Methods:** 110 patients were randomized to either 1 cycle (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, weekly x5). Patients received CT scans at baseline and 3, 6, 9, 12 and 18 months. The primary endpoint was overall survival. **Results:** Final 12-month survival was 32% (35/109), 18-month survival was 22% (24/109) and 24-month survival was 18% (16/91). The response rate was 11% (5 CRs and 6 PRs) with tumor responses observed in both treatment arms; 31 additional patients had stable disease ≥3 months, for a disease control rate of 38% (42/109). Average duration of response in both treatment groups was 9.5 months. Treatment with ADXS11-001 demonstrated patterns of immune response consistent with those seen with other immunotherapies. The addition of cisplatin to ADXS11-001 did not improve survival or tumor response over ADXS11-001 alone. Long-term survivors (LTS) were defined as the 24 patients alive at ≥18 months of which 25% (3/12) were ECOG 2 at baseline and 8% (2/24) had received at least 2 prior treatments for their cervical cancer. 58% (14/24) of LTS had some degree of tumor reduction and 25% (6/24) had tumor burden increases. LTS were evenly distributed between both treatment groups. Baseline ECOG performance status, type of prior therapy, or aggressiveness of disease had no significant effect on survival or tumor response. ADXS11-001 was well tolerated as 62% (68/109) of patients reported no adverse events and 38% (41/109) of patients reported only mild transient adverse events (G1-2) that occurred on the day of infusion. The incidence of SAEs possibly related/ related to ADXS11-001 was 1% G3 (0% G4-5). Activation of innate immunity was demonstrated by increased expression of cytokines (IL6, IL-8, IL-10, INF-γ and TNF-α) and chemokines (MIP-1α, MIP-1β and MCP-1); an increased ratio between ICAM-1/ MIP-1β was associated with infusion related AEs. **Conclusions:** ADXS11-001 appears to be an active agent in patients with recurrent cervical cancer and compares favorably with more toxic treatment options.

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 *Listeria monocytogenes*

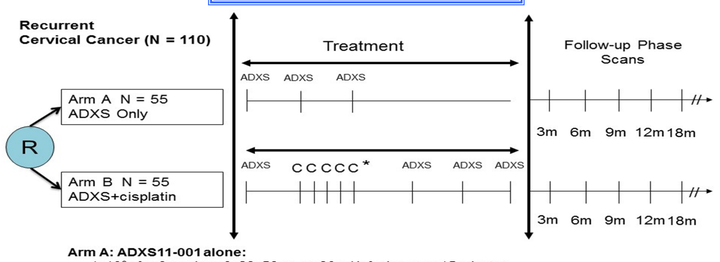
- Attenuation: Genetically Engineered
  - Loss of bacterial virulence due to 10,000 to 100,000 fold attenuation
    - Deletion of Δ *prfA (with D133v complementation)* results in reduction of bacterial virulence factors
    - Recombination and restoration of virulence not possible
  - Secretes HPV-E7 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 100,000 x the LD50 of wild type *Lm* in normal mice.

Live attenuated *Lm* bioengineered to secrete an antigen-adjuvant fusion protein (antigen + tLLO) stimulate a profound innate immune response and are selectively phagocytized by antigen presenting cells (APC). Fragments from *Lm* are processed via the MHC class II 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.

Lm-LLO-E7-15: 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:

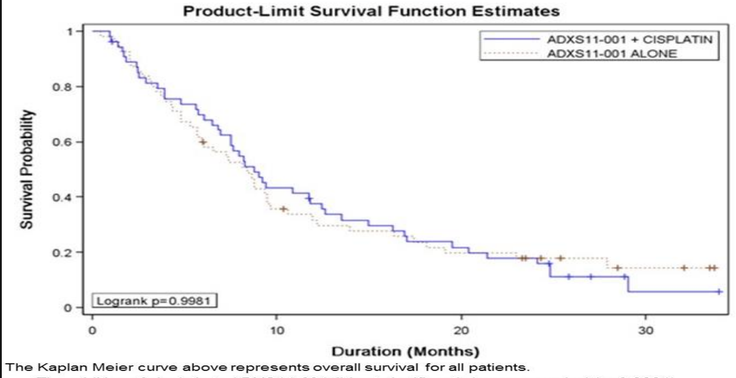
- **41 patients (38%) reported 76 Grade 1-2 AEs**
  - 41 Chills/Shivering
  - 13 Flu Like Symptoms
  - 6 Vomiting
  - 5 Nausea
  - 5 Fever
  - 2 Dizziness
  - 1 Cytokine Release Syndrome
  - 1 Headache
  - 1 Weight Decreased
  - 1 Blood Alkaline Phosphatase Increased
- **1 Grade 3 AE reported as Fever**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



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

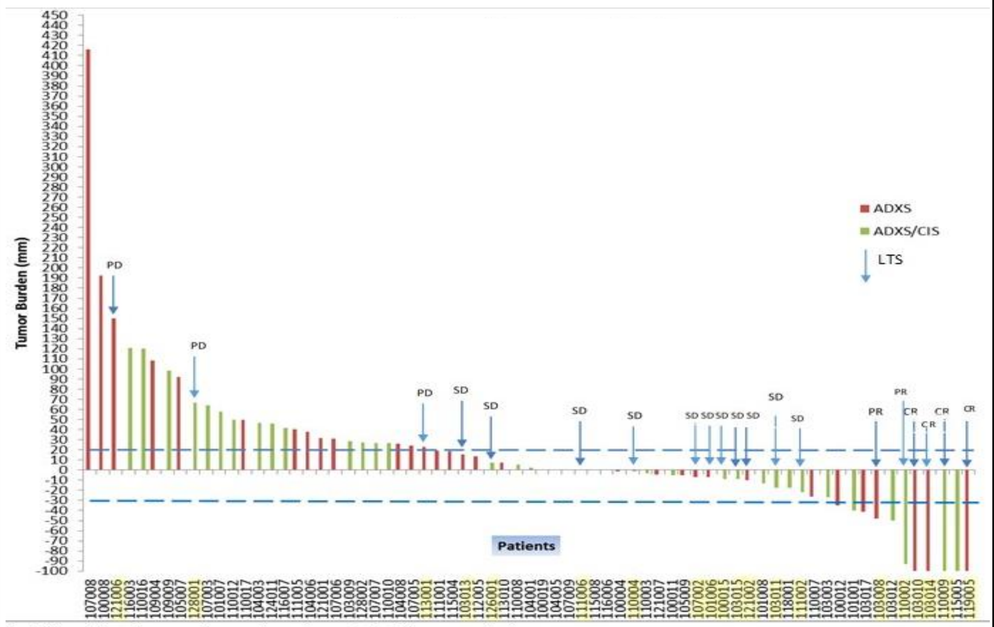
Landmark Survival

Patients at Risk, n (%)*	Overall (N=109)	Treatment Group: ADXS11-001 ALONE (N=55)	Treatment Group: 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	46	45
% alive (#)	18% (16/91)	15% (7/46)	20% (9/45)

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

- 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.

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)

Conclusion

- **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**
  - 10% 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**