**Newsletter**

This resume of the results from the phase 1b study with Foxy-5 is based on clinical and laboratory data from the study, and these data have now been locked into the database. The full report will not be published at this stage, due to pending patent issues and competitor matters.

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### Phase 1b Dose Escalating Study to Evaluate the Safety, Tolerability and Pharmacodynamic response of Foxy-5 in Patients with Metastatic Breast-, Colon- or Prostate Cancer

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**Primary Objective:**
- To evaluate the safety and tolerability of treatment with Foxy-5 defined as presence of Dose Limiting Toxicities (DLTs)

**Secondary Objectives:**
- To determine the maximum tolerated dose (MTD) of Foxy-5 (determined as the dose preceding the dose at which two or more patients have experienced DLTs)
- To characterize the pharmacokinetic (PK) profile of Foxy-5
- To characterize the pharmacodynamic (PD) profile of Foxy-5
- To characterize the pharmacodynamic profile of Foxy-5 to enable the determination of the biological response dose based on the alterations in the following exploratory biomarkers during treatment with Foxy-5:
  - Genome wide mRNA gene expression in tumor biopsies and blood (buffy coat)
  - Wnt-5a protein expression of tumor biopsies
  - Numbers of circulating tumor cells (CTCs) in blood

**OBJECTIVES**

This study was a multi-center, open-label phase 1b dose-escalating study to evaluate the safety, tolerability, dose limiting toxicities (DLT) and maximum tolerated dose (MTD) of Foxy-5 in patients with metastatic breast, colon or prostate cancer for which no approved standard treatment was available. The study was performed in Denmark and the United Kingdom. Additional secondary objectives were to characterize the pharmacokinetic and pharmacodynamic profiles of Foxy-5 as well as determining if a biological response dose could be identified. A biological response was defined as changes in gene expression in tumors and blood (buffy coat), changes in Wnt-5a expression in tumors and estimation of the number of circulating tumor cells before and after treatment with Foxy-5.

**DESIGN**

The study was designed as a standard 3+3 dose escalation cohort design, where a minimum of 3 up to a maximum of 18 patients could be treated at each dose level, and the escalation to the next dose level was permitted depending on the outcomes of the previous dose levels. Escalation to the next dose level was dependent on approval of an independent Data and Safety Monitoring Board (DSMB) that evaluated all relevant safety data for each dose cohort once the last patient in the cohort had completed one cycle of treatment. Cohort expansion to 6 patients was required if 1 DLT was reported. The dose escalation should be stopped if 2 patients experienced a DLT at a given dosage level, and the preceding cohort would then be
expanded to include between 6 and 18 patients, assuming that no DLTs were reported at this dose level, this would be classified as the MTD. The study could also be stopped based on a positive biological response in one of the exploratory pharmacodynamic parameters investigated. The data from exploratory pharmacodynamic parameters was reviewed on an ongoing basis, and data was shared with Investigators as the data emerged. A biological response in any of the pharmacodynamic parameters was classified according to a ‘positive/negative’ principle. If dose escalation was stopped due to an observed biological response in the cohort, this dose cohort could be expanded to include between 6 and 18 patients, and assuming no DLT’s were reported, this dose would be classified as the maximum recommended dose for phase II.

Patients received one intravenous infusion of Foxy-5 three times a week for three weeks; this constituted one treatment cycle. During this cycle the safety and tolerability, pharmacokinetics and pharmacodynamics were investigated. Additional dosing weeks followed immediately after, with no interruption between the dosing weeks. Patients could continue to receive Foxy-5 treatment at the Investigator’s discretion until disease progression or unacceptable toxicity. CT or MRI scans of the patient’s tumor where performed before treatment and after 8 weeks and 12 weeks of treatment and every 8 weeks thereafter until disease progression or patient withdrawal. The patient’s tumor response was assessed according to RECIST criteria (version 1.1). Following disease progression or withdrawal, patients were followed-up for 30 days after End of Treatment (defined as end of all treatment and not end of the first cycle of treatment).

RECRUITMENT OF PATIENTS
A total of 17 patients were included in the study and all patients were included in the evaluation of safety and analysis of biological response. 1 patient at the 1.3 mg/kg dose level did not complete at least 9 treatments (3 weeks) with Foxy-5 and was withdrawn in accordance with the protocol due to progression of disease before completion of the first treatment cycle. All included patients had no or low Wnt5a protein expression in their primary tumors.

TREATMENT
Patients received one intravenous slow infusion of Foxy-5 three times weekly for 3 weeks (1 cycle). Foxy-5 treatment continued at the Investigator’s discretion. The protocol allowed for a total number of 8 dose levels to be tested (0.8mg/kg -7.0 mg/kg). However only 4 dose levels were tested (0.8 mg/kg - 2.3 mg/kg see figure 1), due to dose escalation being stopped as results from exploratory pharmacodynamic parameters suggested that a biological response, displayed by changes in mRNA gene expression in tumor biopsies, was observed at all doses tested.
Figure 1: shows the number of patients who completed each dose level of Foxy-5

As can be seen in Figure 2, many of the patients received additional infusions of Foxy-5. The per protocol number of Foxy-5 infusions were 9 infusions (3 infusions per week for 3 weeks) but the mean number of infusions of Foxy-5 turned out to be 24.1 with three patients receiving 35 or more infusions.

Figure 2: shows the number of infusions with Foxy-5

PRIMARY OBJECTIVE
Toxicity from Foxy-5 treatment was assessed by NCI Common Toxicity Criteria for Adverse Events (CTCAE) version 4.0. The study showed that Foxy-5 was a safe and well tolerated drug at all dose levels and no Dose Limiting Toxicities were observed at any of the doses.

A total of 9 serious adverse events (SAE’s) were observed in this study and the majority of the SAEs were deemed unrelated to treatment. Due to the early stage in clinical development of Foxy-5, all related or possibly related SAEs were to be reported as Suspected, Unexpected, Serious Adverse Reactions (SUSARs). One SAE was classified as possibly related to treatment (oppression of the chest) by the investigator, and was reported as a SUSAR, all others were rated as unrelated to treatment, and none were considered to be dose limiting.

SECONDARY OBJECTIVES
Determination of Maximum Tolerated Dose: The MTD was not determined from this phase Ib study as the study was stopped in accordance with the protocol (positive pharmacodynamics results) when a biological response was observed with all doses of Foxy-5 (0.8 mg/kg, 1.3 mg/kg, 1.8 mg/kg and 2.3 mg/kg). Since no dose limiting toxicities were observed at the dose levels tested, a maximum tolerated dose (MTD) could not be defined.

As part of the secondary objectives the pharmacokinetics and pharmacodynamics of Foxy-5 were also studied.

Pharmacokinetics: Foxy-5 could be detected in the blood, with highest levels either immediately after or 5 minutes after infusion. There was also a rapid initial rate of elimination of 80% of the plasma concentration
with a slower terminal half-life, especially at higher concentrations. This study showed the pharmacokinetics of Foxy-5 to be uncomplicated with dose proportionality and no accumulation.

Pharmacodynamics: According to the protocol, measurements before and during Foxy-5 treatment of the following parameters were used to estimate Foxy-5 pharmacodynamics:

1. mRNA expression in tumors and in blood (buffy coat) before and during Foxy-5 treatment
2. Wnt-5a expression in tumor biopsies before and during Foxy-5 treatment
3. Circulating tumor cells before and during Foxy-5 treatment

CT or MRI scans were also performed before, during and after Foxy-5 treatment as a secondary effect endpoint, however the results were for information only and the study could not be stopped based positive results from this evaluation, and as such results have not been included in this description.

mRNA expression in tumors and blood (buffy coat) before and during Foxy-5 treatment: Tumor biopsies (one biopsy before and one after administration of Foxy-5) and blood samples were obtained from 15 patients enrolled in this phase 1b study. In addition to this, biopsy material and blood samples from 3 patients from the highest dose level (1.3 mg/kg) in the first phase 1 study was also available for analysis. The data set therefore contained sample material from a total of 18 patients across the phase 1 program. These 18 patients consisted of 14 males and 4 females, 10 had colon cancer, 6 had prostate cancer and 2 had breast cancer. The group of patients was very diverse in terms of gender and type of cancer and also the number of patients for each dose level was limited. Because of this diversity, no formal statistical analysis could be used to evaluate the data, and thus each patient has been evaluated separately within each dose level. For each patient, the mean value of the 5, 10 and 20 genes with the highest fold change – both regarding up-regulation and down-regulation - were plotted to evaluate if a biological response had been obtained.

The results from buffy coat analysis did not reveal any systematical changes in gene expression during or after treatment. As can be seen from the figure below, regardless if one analyzed the 5, 10 or 20 most up-regulated or down-regulated genes no response was even close to a 2-fold change in up- or down regulation of gene expression, which were the predefined criteria for a positive response in gene regulation.
Figure 3: (Dose 1 = 0.8 mg/kg, dose 2 = 1.3 mg/kg, dose 3 = 1.8 mg/kg and dose 4 = 2.3 mg/kg - Each point represents the mean of the top 5, 10 or 20 gene fold change, while the bar-plots represent the median of the points (median of the fold change means).
For the tumor biopsies, the results were very positive and encouraging and showed that a change in gene expression could be observed after treatment for all dose levels tested. The greatest change in up-regulated gene expression was observed for the 1.8 mg/kg dose level (dose level 3), where an approximately 3.0- to 3.5-fold change was observed across the top-most regulated genes. The greatest change in down-regulated gene expression occurred at the 1.3 mg/kg dose level (cohort 2) and was maintained at the 1.8mg/kg dose level and was approximately 2.1 to 2.5 across the top-most regulated genes.
**Figure 4:** Dose 1 = 0.8 mg/kg, dose 2 = 1.3 mg/kg, dose 3 = 1.8 mg/kg and dose 4 = 2.3 mg/kg. Each point represents the mean of the top 5, 10 or 20 most regulated genes and is presented as the mean fold change for each patient. The bar-plots represent the median of the points (median of the fold change means).

Based on these results, it was concluded that all doses tested so far in the study had resulted in a positive biological response directly in the patient’s tumors and the study was therefore stopped in accordance with the protocol. It is important to notice that at this point it is not possible to say that the observed changes in gene expression are related to the metastatic cancer process, but our interpretation is that Foxy-5 reaches its target and that a biological response to this is observed.

Circulating tumor cells before and during Foxy-5 treatment: Measurements of circulating tumor cells in the blood was obtained from all patients in this study. Two different methods (CellSearch and Pasortix) were used to analyse samples and especially the CellSearch method showed good comparability in measurements between 2 simultaneously collected blood samples from the patient. Results from this method are depicted in Figure 5, and all patients except 1 had detectable cancer cells in the blood. Results did not reveal any apparent systematic or consistent changes across dose levels or type of cancer.

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**Figure 5:** Number of CTC (circulating cancer cells) at day 1 before treatment, day 10/12 and day 17/19 after Foxy-5 treatment.
Wnt-5a tumor cell content before and during Foxy-5 treatment: Wnt-5a expression analysis was performed on tumor biopsies taken from metastatic cancer tissue (one biopsy before and one after end of the Foxy-5 treatment). Material was obtained from 18 patients enrolled in this phase 1b study, however 3 samples were of poor quality and therefore only samples from 15 patients could be used. In addition to this, biopsy material from 3 patients from the highest dose level (1.3 mg/kg) in the first phase 1 study was also available for analysis. The data set therefore contained sample material from a total of 18 patients across the phase I program.

A comparison between the two biopsies taken from each patient revealed that the majority of biopsy pairs (n=11) showed no difference in Wnt-5a protein expression following end of treatment with Foxy-5. The data is limited and it is not possible to draw any conclusions regarding any effect of Foxy-5. It is very obvious when evaluating the complete tumor sections from primary tumors that it consists of areas that are Wnt-5a positive and areas that are Wnt-5a negative. In this study, classification of a patient’s tumor as “Wnt-5a negative” was when the tumor sample was predominantly (> 50%) Wnt-5a negative and as a tumor biopsy only represents a minor part of the tumor tissue there is a risk that two biopsies taken from the same tumor can show different expressions of Wnt-5a and that these changes are not related to a specific change, but rather due to a heterogenous expression of Wnt-5a in tumor tissue.

DISCUSSION AND CONCLUSIONS
The planed clinical phase 1 study, with 3 patients at dose level 1 and 4, 4 patients at dose level 2 and and 7 patients at dose level 3, has been finalized.

The main finding is that Foxy-5 is safe and well tolerated and that no dose limiting toxicity was found at the dose levels applied (0.8 mg/kg to 2.3 mg/kg). The MTD was not determined as the study was stopped in accordance with the protocol, when positive pharmacodynamics results were obtained, and a biological response was observed. A biological response in gene expression (mRNA) from tumor biopsies was observed with all doses of Foxy-5 (0.8 mg/kg, 1.3 mg/kg, 1.8 mg/kg and 2.3 mg/kg); these exploratory results were obtained for all doses simultaneously after which the study was stopped (3 patients from 4 dose levels had been treated with Foxy-5 at this time due to parallel initiation of dosing in dose level 1 and 3). Dose escalation therefore did not continue after dose level 4 (2.3 mg/kg). Based on the preliminary results available, it was decided to expand dose level 3 (1.8 mg/kg) to include 7 patients. Dose level 3 was identified as the most suitable dose level for expansion when considering the available pharmacodynamic results, available safety data from non-clinical toxicity studies as well as clinical safety data and finally also pharmaceutical aspects. As the study was stopped before the final planned dose level was reached, the MTD may have been observed if dose escalation had continued past dose level 4.

The finding that Foxy-5 produces a biological response in the dose levels tested is very positive as it not only indicates that Foxy-5 reaches its intended target and has an effect, but also provides a foundation for selection of the most optimal dose for future phase II studies.

In the buffy coat analysis, no systematic changes in gene expression during Foxy-5 treatment were observed. This is in line with the results from the phase 1 study, where no Foxy-5 induced toxicities from any organ were noted and also no signs of Foxy-5 effects were observed in blood samples collected for safety assessment. Moreover, examinations of organs from rats and dogs dosed with Foxy-5 in the non-clinical studies did not show any changes from normal after Foxy-5 treatment. The results from buffy coat analysis during this study suggest that Foxy-5 treatment does not affect gene expression from circulating blood/normal (non-tumor) cells and therefore buffy coat analysis could possibly be used as a baseline control for gene expression studies in tumor biopsies, but further analysis and standardization of methods are required to establish if this is possible.
Measuring CTC before and during Foxy-5 treatment did not reveal any systematic changes in CTC numbers. CTC counts were still difficult to evaluate even though new approaches / methods were employed. As previously described, the reason for this may be because Foxy-5 impairs at least two different processes, the release of tumor cells from tumor tissues (decreasing the number of CTC’s) and the passage of cancer cells out of the blood stream (increasing the number of CTC’s).

As per protocol, we performed CT scans every 8 weeks following treatment initiation. We did observe three patients with a confirmed stable disease as determined by CT or MRI scans. Due to the limited number of patients in this study and the lack of an untreated control group, we are not able to make any firm conclusions on this finding.

The safety data from this study are in line with the previously performed phase I study and combined with the safe profile of Foxy-5 observed from animal toxicology studies, and especially the positive pharmacodynamic results obtained in the phase Ib study, the company finds that the accumulated data presents a positive benefit/risk relationship for Foxy-5 for the current stage in development and will form a solid basis for evaluating the recommended dose for the subsequent phase II study and later clinical stage development.