Formulier
Projectvoorstel dierproeven

- Dit formulier gebruikt u om uw projectvoorstel van de dierproeven te schrijven
- Bij dit formulier hoort de bijlage Beschrijving dierproeven. Per type dierproef moet u deze bijlage toevoegen.
- Meer informatie vindt u in de *'Toelichting op de te gebruiken formulieren voor de aanvraag van een projectvergunning'* op de website www.centralecommissiedierproeven.nl.
- Of neem telefonisch contact op. (0900-2800028).

1 Algemene gegevens

1.1	Vul uw deelnemernummer van de NVWA in.	[redacted]
1.2	Vul de naam van de instelling of organisatie in.	Biomedical Primate Research Centre
1.3	Vul de titel van het project in.	Evaluation of safety and immunogenicity in rhesus macaques of a therapeutic vaccine for treatment of HER2 positive tumours in humans

2 Categorie van het project

2.1 In welke categorie valt het project?

U kunt meerdere mogelijkheden kiezen.

- Fundamenteel onderzoek
- Translationeel of toegepast onderzoek
- Wettelijk vereist onderzoek of routinematige productie
- Onderzoek ter bescherming van het milieu in het belang van de gezondheid of het welzijn van mens of dier
- Onderzoek gericht op het behoud van de diersoort
- Hoger onderwijs of opleiding
- Forensisch onderzoek
- Instandhouding van kolonies van genetisch gemodificeerde dieren, niet gebruikt in andere dierproeven

3 Algemene projectbeschrijving

3.1 Achtergrond

Licht het project toe. Beschrijf de aanleiding, de achtergrond en de context. Besteed aandacht aan de bij vraag 2.1 aangekruiste categorieën.

Worldwide, breast cancer is the leading type of cancer in women, accounting for 25% of all cases[1]. Breast cancer is a widespread oncology indication affecting more than 2 million people worldwide annually and resulting in more than 450,000 deaths[2]. Approximately 20-30% of all breast cancers overexpress the human epidermal growth factor receptor 2 [3-8]. HER2-positivity has historically conferred a poor prognosis

owing to its association with high-grade histology, lymph node involvement, and higher rates of disease recurrence and mortality[3-8]. HER2 is a member of the human epidermal growth factor receptor (HER/EGFR/ERBB) family. Amplification or over-expression of this oncogene has been shown to play an important role in the development and progression of certain aggressive types of breast cancer. In addition to breast cancer, HER2 over-expression is also known to occur in ovarian[9], stomach, adenocarcinoma of the lung[9] and aggressive forms of uterine cancer, such as uterine serous endometrial carcinoma[10,11], e.g. HER2 is over-expressed in approximately 7-34% of patients with gastric cancer[12,13] and in 30% of salivary duct carcinomas[14]. In recent years the protein has become an important biomarker and target of therapy for approximately 20-30% of breast cancer patients[8].

The monoclonal antibody (mAb) trastuzumab, that binds to HER2 and was developed over 20 years ago, represented a revolutionary improvement in the treatment of HER-2-positive breast cancer[15]. Anti-HER2 agents have dramatically improved patient outcomes, when applied before or after the main treatment, for early disease, and similarly in advanced or metastatic disease through first- and later-line application. Currently several anti-HER2 agents are in clinical use including the anti-HER2 monoclonal antibodies trastuzumab and pertuzumab; the small-molecule inhibitors lapatinib, neratinib, and tucatinib; and the antibody–chemotherapeutic drug conjugates ado-trastuzumab emtansine (TDM1) and trastuzumab deruxtecan. In addition, a number of trastuzumab biosimilars have recently been granted regulatory approval in North America and Europe and are enhancing patient access to targeted therapies[16].

Treatment of breast cancer by targeting HER2 is effective, however the use of mAb therapy has several drawbacks. Firstly, the treatment is very expensive. Secondly, administration of large doses of mAb (despite being "humanised") is associated with severe side effects due to the development of anti-mAb immunity and these anti-mAb antibodies subsequently reduce the efficacy of treatment. Thirdly, resistance against the mAb therapy develops because of mutations in the native HER2 antigen, thereby compromising treatment efficacy.

These limitations have prompted the development of anti-HER2 vaccines capable of triggering the patient's own immune system to produce anti-tumour antibodies (Ab)[17-20]. In this regard, the main hurdle has been to generate robust and durable anti-tumour immune responses. Monomeric proteins are generally weak immunogens and simple subunit vaccines based on a soluble protein antigen in an adjuvant formulation have almost exclusively failed in clinical trials due to insufficient efficacy. In the context of anti-cancer vaccines multiple mechanisms prevent induction of immune responses against self-antigens. Central T-cell tolerance prevents the egress of auto-reactive T cells from the thymus and the tumour environment imposes additional immune tolerogenic mechanisms to prevent induction of $CD4^+$ T-helper cells[21]. Consequently, auto-reactive B cells, which can be found in the circulation[22], do not receive co-stimulation from $CD4^+$ T-helper cells, which is required for their full activation and subsequent affinity maturation and isotype-switch of the Ab[23]. Contrary to monomeric proteins, surface of virus-like particles (VLPs) are highly immunogenic due to sharing key characteristics with live viruses. Their repetitive surface structures facilitate complement fixation and B cell receptor clustering, which activate the innate immune system and leads to greater B cell activation[24,25]. Multivalent display (e.g. on the surface of a VLP) can moreover abrogate the ability of the humoral immune system to distinguish between self and foreign and promote class switching, multiplication, affinity maturation, and survival of autoreactive B cells[26,27]. Consequently, VLP-display of a self-antigen is critical for inducing strong and durable autoantibody responses[27].

A recently developed VLP-HER2 vaccine, with directional high-density display of the human HER2 ectodomain on the surface of a VLP was shown to be safe and efficacious in murine models of breast cancer; Self -tolerance was broken in huHER2 transgenic mice[28] and Ab induced by the VLP-HER2 vaccine were shown to be efficacious in murine tumour models[28]. A therapeutic breast cancer vaccine based on VLP-HER2 can potentially ameliorate several of the limitations associated with mAb therapy. Costs of repeated (booster) immunisations are reduced as compared to repeated mAb treatment. Repeated immunisations with the VLP-HER2 vaccine will not induce detrimental anti-Ab immunity. In addition, the immune response induced by the VLP-HER2 vaccine is polyclonal and targets multiple epitopes on the HER2 antigen, thereby reducing the chance of HER2 escape mutations. The rhesus macaque has a HER2 extracellular domain that is 99% homologous to the human HER2 protein[29], underscoring the relevance of the rhesus macaque for this type of experiments, where self-tolerance needs to be broken.

3.2 Doel

3.2.1 Beschrijf het directe en het uiteindelijke doel van het project. Beschrijf de bijdrage van het behalen van het directe doel aan het uiteindelijke doel.

- Indien het directe doel bestaat uit verschillende subdoelstellingen, benoem deze dan hier.

The direct aim of this proposal is to evaluate the VLP-HER2 vaccine for occurrence of adverse effects and immunogenicity in rhesus macaques. The ultimate aim of this proposal is to develop a therapeutic VLP-HER2 vaccine for patients with HER2 positive tumours.

3.2.2 Hoe wordt de haalbaarheid van het directe doel gewaarborgd?

At our institute we have been performing vaccine evaluation studies in NHP for over 20 years. Many previously evaluated vaccines were directed against human immunodeficiency virus, hepatitis B virus, hepatitis C virus, malaria, influenza and tuberculosis. In addition, we have previously evaluated safety and immunogenicity of a vaccine directed against the p53 oncogene. We have the appropriate immunological assays for assessment of cellular, humoral and innate immune responses against vaccine antigens. Our long-standing experience with vaccine evaluation guarantees that the animal studies described in this proposal will be adequately performed.

3.2.3 Is voor de uitvoering van dit project andere wet- en regelgeving van toepassing die een invloed zou kunnen hebben op het welzijn van de dieren en/of de haalbaarheid van het directe doel?

Nee

Ja > Geef aan welke wet-en regelgeving van toepassing is en beschrijf de effecten daarvan op het welzijn van de dieren en de haalbaarheid van het project.

3.3 Belang

3.3.1 Beschrijf het wetenschappelijk en/of maatschappelijk belang van de hierboven beschreven doelen.

Worldwide, HER2 positive tumours account for about 20-30% of breast cancers. Although treatment is available in the form of mAb therapies, a therapeutic vaccine, with advantages as outlined under 3.1., offers a new potentially better therapeutic option for patients with HER2 positive tumours. In the event the vaccine has shown to be safe and immunogenic in this project, the VLP-HER2 vaccine will be evaluated in clinical trials.

3.3.2 Benoem de belanghebbenden in het project en beschrijf voor elk van de belanghebbenden wat hun belang is.

Rhesus macaques do not suffer from HER2-positive tumours, therefore the evaluated vaccine does not offer any benefits for the animals. The animals will experience moderate discomfort due to repeated sedations, vaccinations and blood collections.
Patients with HER2-positive tumours are stakeholders for a HER2 vaccine, for whom a new therapeutic option may become available. This would also be of societal benefit as treatment complexity and costs will be reduced.
The proof of concept for the VLP-HER2 vaccine will also impact vaccine science and can, if proven successful, also be applied for other (tumour) antigens.
If the vaccine proves to be safe and immunogenic in rhesus macaques the manufacturer of the vaccine can proceed to clinical trials. When a product becomes available following completion of the clinical trials, the manufacturer can financially benefit from their intellectual property rights on the VLP-HER2 vaccine.

3.4 Strategie

3.4.1 Geef een overzicht van de algemene opzet van het project. Besteed aandacht aan de eventuele fasering in de uitvoering en de samenhang. Vermeld eventuele mijlpalen, keuzemomenten en besliscriteria.

In order to evaluate the safety and immunogenicity of the vaccine concept, a vaccine evaluation experiment will be performed according to well established procedures, as described in appendix 1. Typically, a number of immunisations are given over a certain time period. Following immunisation induction of systemic T-cell

and antibody immune responses are measured, in the blood. The strength of these responses as well as their duration are determined. Vaccine-induced Ab levels will be compared to therapeutic levels of mAb (ranging between 80 - 115 µg/mL)[30] to evaluate the clinical potential of the vaccine. The criteria to consider vaccine candidates for evaluation are: a) the vaccine strategy must be novel, for instance with regards to choice of antigen, formulation, route of application, that have not been tested before in similar NHPs studies, b) demonstration that the vaccine or vaccine components are non-toxic, c) cross recognition of macaque homologues must have been demonstrated, d) the vaccine cannot be adequately tested in other than NHP animal models, due to required similarity between human HER2 and HER2 in the test species or the type of immunological assessment needed, e) preferably immunogenicity of vaccine candidates should have been proven in other species, unless this is not possible because the specific vaccine modality used does not work in other species.

3.4.2 Onderbouw de gekozen strategie.

For this type of experiment animals will be immunised several times (max 5) over a defined time period. During the study animals will be monitored for adverse effects of the vaccine, including monitoring of injection sites, general behaviour and health and systemically measuring haematology and clinical chemistry parameters. Blood will be collected to determine induction of systemic immune responses.

3.4.3 Benoem de type dierproeven. Vul per type dierproef een bijlage Beschrijving dierproeven in.

Volgnummer	Titel bijlage Beschrijving dierproef
1	Safety and immunogenicity evaluation of a therapeutic vaccine for HER2 positive tumours

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