Study Objectives

1. Assess the diagnostic sensitivity and specificity of [^{68}Ga]Ga-DOTA-mDesmo PET/CT in identifying corticotropinomas.
2. Evaluate the accuracy of [^{68}Ga]Ga-DOTA-mDesmo PET/CT in localizing tumors compared to MRI and BIPSS.
3. Determine the impact of [^{68}Ga]Ga-DOTA-mDesmo PET/CT on surgical outcomes, including remission rates post-transsphenoidal surgery.

Study Protocol

1. **Biochemical Tests**

Serum cortisol dynamics (11 am and 8 pm) and ACTH will be measured by electro-chemiluminescence-immuno-assay (ECLIA) (ELECSYS-2010, Roche Diagnostics, Germany).

2. **Bilateral inferior petrosal sinus sampling (BIPSS)**

BIPSS will be performed in patients: 1) wherever possible, and 2) in cases where confusion between the diagnosis of Cushing's disease and ectopic Cushing's disease will arise.

3. **Contrast-Enhanced Magnetic Resonance Imaging** (**CE-MRI**)

Dynamic CE-MRI of the sella with gadolinium-enhanced spoiled gradient (SPGR) T1 imaging will be conducted on a 3T MRI scanner. CE-MRI will be performed in two scenarios: 1) on confirmed cases of Cushing's disease to localize and lateralize the pituitary adenoma for trans-sphenoidal surgery, and 2) in cases where confusion regarding the ACTH source between the pituitary source and ectopic sources may arise.

4. **[^{68}Ga]Ga-DOTA-mDesmo PET/CT Patient Imaging**

Inclusion Criteria

i. Patients aged 8 years and older.
ii. Clinically diagnosed with Cushing's disease.
iii. Patients scheduled for imaging studies to localize corticotropinomas.
iv. Informed consent.

Exclusion Criteria

i. Patients with exogenous exposure to steroids

ii. Pregnancy or lactation.

iii. Severe comorbid conditions that may interfere with the study.

Imaging with [^{68}Ga]Ga-DOTA-mDesmo will be performed after obtaining Institutional Ethics Committee approval and written informed consent from the patients. Patients will be monitored for any adverse events during and after 2 hours of [^{68}Ga]Ga-DOTA-mDesmo injection. Several vital parameters will be recorded, such as blood pressure, body temperature, heart rate, and oxygen saturation levels. The patients will undergo regional brain PET/CT imaging post intravenous administration of [^{68}Ga]Ga-DOTA-mDesmo (72-148 MBq) using a hybrid 3D PET/CT scanner (Siemens Healthineers Biograph mCT S-64, Germany). The regional CT acquisition will be performed with a tube voltage of 120 keV; a tube current of 100-350 mA (Auto mA) and a slice thickness of 0.6 mm. The CT images will be acquired for the brain region without intravenous contrast media, followed by a regional brain PET scan with an acquisition time of 10 mins. The CT data will be used for attenuation correction of PET images. The PET images will be reconstructed in a matrix of 256 x 256 using an ordered subset expectation maximization (OSEM) algorithm (21 subsets, 2 iterations) and a z-axis Gaussian filter with FWHM of 5 mm. The two experienced nuclear medicine physicians who will be unaware of the patient's history will interpret the PET/CT images. A region of interest (ROI) will be drawn around the abnormal uptake of [^{68}Ga]Ga-DOTA-mDesmo. The lesion activity will be represented as the maximum standardized uptake value (SUV_{max}). The SUV_{max} values of the lesion will be compared with the surrounding healthy tissue (background SUV_{max}). CEMRI images, where available, will be superimposed with the [^{68}Ga]Ga-DOTA-mDesmo PET/CT images during analysis.

5. **Histopathology**

The lesions detected on either CE-MRI or [^{68}Ga]Ga-DOTA-mDesmo PET/CT will undergo transsphenoidal surgery. The surgical tissues will be examined for ACTH immunostaining positivity. The diagnosis of Cushing's disease will be confirmed only if histopathology shows evidence of corticotropinoma. The outcomes of histopathology will serve as a gold standard.

6. Data Collection

Data collection will occur in two phases: preoperative and postoperative.

Preoperative Data Collection:

1. **Clinical Characteristics**: Collect demographic information (age, sex), medical history, and clinical symptoms related to Cushing's syndrome.
2. **Imaging Results**: Document the findings from CE-MRI and [^{68}Ga]Ga-DOTA-mDesmo PET/CT, including tumor localization, size, and any metastases.
3. **Tumor Characteristics**: Record tumor size (measured in mm), location (e.g., sella turcica), and any relevant imaging characteristics that may influence surgical planning.

Postoperative Data Collection:

1. **Histopathological Assessments:** The surgical tissues will be examined for ACTH immunostaining positivity.
2. **Remission Rates**: Defined by biochemical markers such as serum ACTH levels and cortisol levels measured at regular intervals (e.g., 3-months, 6-months, 1-year post-surgery).
3. **Clinical Assessments**: Follow-up visits will be conducted to assess clinical symptoms and overall health.
4. **Complications**: Document any surgical complications or adverse events that occur during hospitalization or follow-up.

Follow-Up Period

We will establish a defined follow-up period for assessing remission rates and overall outcomes:

1. **Short-Term Follow-Up**: Initial assessments at 3 months post-surgery to evaluate early remission rates and complications.
2. **Medium-Term Follow-Up**: Additional assessments at 6 months to monitor ongoing recovery and biochemical markers.
3. **Long-Term Follow-Up**: A final assessment at 1-year post-surgery to evaluate sustained remission rates and overall patient well-being.

7.0 Statistical Analysis

SPSS will be used for statistical analysis. Descriptive statistics, such as frequencies, proportions, and means will be calculated. Continuous data variable will be represented using the median and Interquartile range (IQR). A manual case-by-case comparison will be conducted to calculate the concordance for localizing and lateralizing the ACTH source. Histopathological outcomes will serve as the gold standard for determining the diagnostic accuracy of [^{68}Ga]Ga-DOTA-mDesmo PET/CT, BIPSS, and MRI. The diagnostic accuracy of [^{68}Ga]Ga-DOTA-mDesmo PET/CT will be compared with that of BIPSS and MRI.

Participant Information Sheet (PIS)

PROTOCOL No:

SPONSOR:

PRINCIPAL INVESTIGATOR: Dr Jaya Shukla

Name of Participant: _____

Title: **mDesmo cold-kit for PET imaging to localize ACTH dependent Cushing syndrome**

1. You are invited to take part in this research study. The information in this document is meant to help you decide whether or not to take part. Please feel free to ask if you have any queries or concerns.

2. **What is your expected duration of the participation?**

   Your participation in this study will last for a total of one day for diagnostic scan **What procedures will be followed during this study?**

   i. Patients will be referred by their treating physicians

   ii. Recent hemogram and blood chemistry reports will be required.

   iii. Informed consent will be obtained before injection.

   iv. An intravenous injection of 2-5 mCi ^{68}Ga-mDesmo will be intravenously injected and PET/CT imaging will be done at 1hr (and 2hr if required) post injection in the Department of Nuclear Medicine. It will be one day procedure.

   v. No hospitalization is required.

4. **What are the risks and discomforts to you?**

   No foreseeable risks are expected. The diagnostic and therapeutic dose will be administered by intravenous route as one time procedure.

5. **What benefits are expected from this research?**

   This is a unique study. The study will address the patients who do not respond to available therapies. Initially we'll perform a diagnostic scan and if the uptake of the drug is visualized, patient may be selected for the therapy after consultation with the treating physician.

6. **What are the alternatives available to you?**

   No alternate therapy is available with us.

7. **Are the data/records of the participant kept confidential?**

a) All the information related to you will be kept confidential as allowed by the applicable laws. Information about your participation in the study will be associated with you by using subject identification number. Only the study doctor or his/her study staff will be able to link that

number to you at all times. Members of ethics committee or regulators designated for auditing the study may see your data if needed. Any publication made out of research study will not identify any study person and only anonymized information and or summaries will be published.

b) The information about your health will be collected and analysed by the study team. These people will have access to your medical information and see your name, other personal information such as year of birth and gender, but will be obliged to keep this information confidential unless required by law or a regulatory authority.

c) You have a right access to your information and the right to correct information but cannot be given information about results from study procedures while the study is being conducted.

8. **What will be the treatment schedule(s)?**

Treatment will be decided based on the diagnostic scan which will be of single day procedure. The treatment protocol as given in point No-3 will be followed.

9. **What compensation and/or treatment(s) are available to the Participant in the event of a trial-related injury?**

This is a diagnostic scan with very small dose of radiopharmaceutical (2-5 mCi). The procedure is similar to any other PET Scan routinely done at the department of Nuclear Medicine. No procedure related injuries are expected/anticipated. However, in case of any unexpected SAE, compensation will be given as per Govt of India rules.

10. **Whom to contact for trial related queries and what are the rights of Participants in the event of any injury?**

For trial related queries and Rights of Participants, you can call persons mentioned as 'Contact persons 'at point no-18. The procedure is non-invasive, and no procedure related injury is anticipated.

11. **Are the participants paid to take part in this study?**

No

12. **What are your responsibilities during participation in the study?**

Based on your type of study (as mentioned in Point No.3), you will be called for one day for diagnostic imaging.

The participation of patient in the study is voluntary. The Participant is free to withdraw from the study at any time. He/ She can also refuse to participate in the study. No penalty will be levied. There will be no loss of benefits to which the Participant is otherwise entitled.

13. The Participant or Participant's representative will be notified in a timely manner if significant new findings develop during the course of the research which may affect the Participant's willingness to continue participation will be provided.

14. A statement that the particular treatment or procedure may involve risks to the embryo or foetus. Therefore, the procedure will not be performed in pregnant participants.

15. Approximate number of Participants enrolled in the study

    Fifty participants will be enrolled in the study

16. Any other pertinent information-Nil

17. **Contact persons:**

    For further information / questions, you can contact us at the following address:

Dr Jaya Shukla Dept. of Nuclear Medicine PGIMER Chandigarh Tel: 7087009143	Dr. Rama Walia Dept. of Endocrinology PGIMER Chandigarh Tel: 91-172-2754757

Convener

In case of conflicts, you can contact the convener of our institutional ethics committee at the following address:

Prof. Ashish Kakkar, Convener, Institutional Ethics Committee, PGIMER, Chandigarh, Telephone: 91-172-2755266

Informed Consent Form (ICF)

Study Title: **mDesmo cold-kit for PET imaging to localize ACTH dependent Cushing syndrome**
Study Number (if any):

Subject's Initials: _____

Subject's Name:_____

Date of Birth / Age: _____

	Participant's initial
1. I confirm that I have read and understood the information sheet dated _ _ _ _ for the above study and have had the opportunity to ask questions.	
2. I understand that my participation in the study is voluntary and that I am free to withdraw at any time, without giving any reason, without my medical care or legal rights being affected.	
3. I understand that the Sponsor of the clinical trial, others working on the Sponsor's behalf, the Ethics Committee and the regulatory authorities will not need my permission to look at my health records both in respect of the current study and any further research that may be conducted in relation to it, even if I withdraw from the trial. I agree to this access. However, I understand that my identity will not be revealed in any information released to third parties or published.	
4. I agree not to restrict the use of any data or results that arise from this study provided such a use is only for scientific purpose(s)	
5. I agree to take part in the above study	

Signature (or Thumb impression) of the Subject/Legally Acceptable Representative:_____

Date: _____/_____/_____ Signatory's Name

_____

Signature of the Investigator: _____
Date:_____/__/_____ Study Investigator's Name

_____