**Engineering a Real-Time Living Biosensor: DNA Damage Caused by Ultra-Violet Irradiation**

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**Motivation**
- 1 million Americans are diagnosed with skin cancer every year.¹
- Current products measure UV radiation level, not DNA damage.
- Our goal: create a real-time sensor of DNA damage

**Approach**
- "If-Then" Construct: Promoter + reporter
  - Promoter: recA of SOS system, part J22106 (activated for extreme DNA damage)
  - Reporter: lacZ, part J732017 (blue/white screening on X-gal)
- If DNA is damaged extensively, then transcription of β-gal
- Essentially a reporter-gene assay

**Mechanisms**
- **SOS Repair Mechanism**
  - SOS induction: UV exposure -> SOS signal
    - SOS genes: recA (Thymine dimer repair), X-gal (β-galactosidase)
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    - SOS genes: recA (Thymine dimer repair), X-gal (β-galactosidase)

**Standard Assembly of the Part**
- Received strain of Top 10 cells from iGEM:
  - LacZ (J732017) and recA promoter (J22106)
- Parts on standard pSB1A2 plasmids (AmpR)
- Grow up to make glycerol stocks
- Miniprep/Digest to check successful assembly

**Building and Testing the System**
- Ligulation with T4 ligase to create engineered plasmid
- Transformation of clone into lac-compliant cells
- Plate on Ampicillin/X-gal
- Dose with UV light to test abilities
- Miniprep/Digest to check successful assembly

**Modeling the SOS System**

**Predicted Response Time: Tens of Minutes**

**Next Steps**
- Perform additional experiments comparing UV radiation to SOS signal
- Compare and refine theoretical model
- Perform experiments with bacteria under UV with different SPF levels
- Develop a sustainable bacterial gel with a shelf life of >3 months
- Create a bio-sensor patch

**Safety**
- Negligible safety issues with project:
  - Pre-engineered E. coli cannot sustain a population outside lab environment
  - Biological Safety Level I only (low risk)
- Project reviewed and approved by the Institutional Biosafety Committee (IBC)
- IBC oversees rDNA research at Purdue
- No characteristics of protocol are considered hazardous

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References