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Rapid Communication

Soluble polymer supported organic synthesis (SPSOS): Synthesis of PEG-ylated phthalizinediones

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PEG-ylated phthalizine derivatives of different molecular weights have been synthesized after the activation of PEGs using glutaric anhydride. Percentage loading has been calculated using ^{1}H NMR. The low molecular weight and disubstituted PEGs have been found to be more reactive as compared to the high molecular weight and tetra substituted PEGs in terms of percentage loading.

Keywords: polyethylene glycol (PEG), pegylation, phthalizinedione

IPC: **Int.Cl.**[7] **C 07 D**

Polyethylene glycols (PEGs), which are hydrophilic and non-toxic, have a variety of biotechnical and biomedical applications including aqueous two phase partitioning[1,2], protein immobilization[3,4] (*i.e.* enzyme, antibody, antigen), drug modification[5] and peptide rejecting surfaces[6].

Recent advances in the high throughput screening[7] of compounds for efficacy in biological assays have revolutionized the drug discovery process. To cater the increasing demand for new molecules, PEGs are exploited extensively in the design and synthesis of bioactive molecules; the efficacy of these modified compounds is often dependent on the length of the PEG chains attached. Nevertheless, the syntheses of long mono disperse PEG chains are rare and remain a synthetic challenge.

Polyethylene glycol is known to enhance water solubility and reduce immunogenicity of high molecular weight protein adducts[8a-c] and therefore seemed like an excellent candidate for conjugation to various carboxylic acid anhydrides. Phthalizine derivatives have numerous applications in biology[9] and synthetic organic chemistry[10,11]. A few methods have been reported[12] in the literature for the synthesis of phthalizine, and their derivatives. However, a suitable methodology is yet to be reported for the synthesis of PEG-conjugated anhydrides.

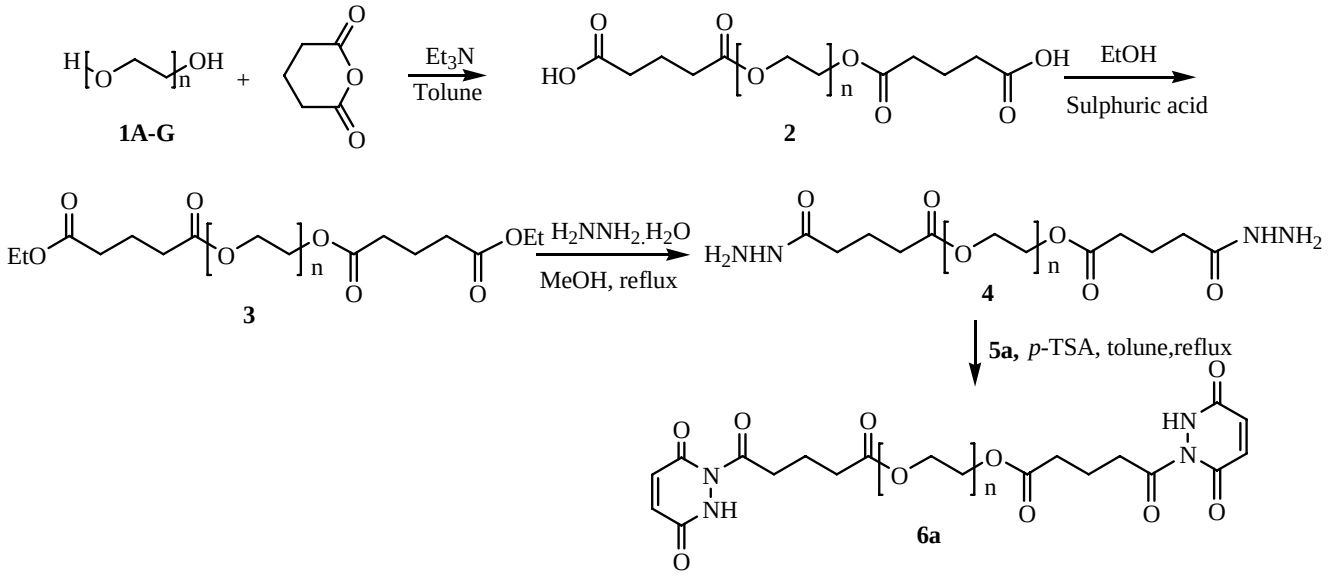
Results and Discussion

Syntheses of PEG-ylated phthalizine diones were carried out after the activation of polyethylene glycol **1A-F** and polyethylene glycol methyl ether **1G** using glutaric anhydride to obtain the respective acids **2A-G**. These PEG-ylated acids were converted to their respective ethyl esters using absolute ethanol and catalytic amount of sulphuric acid. These esters **3A-G** were treated with hydrazine hydrate to obtain hydrazides **4A-G**. The hydrazides were subjected to cyclization using different anhydrides **5a-e** in the presence of *p*-TSA to get the final PEG-ylated phthalizine diones. Though *p*-TSA was widely used in several organic transformations, there is no report for its use in the condensation of PEG with anhydrides to give PEG-bound phthalizine derivatives (**Scheme I**).

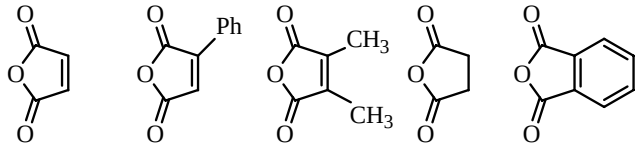
Experimental Section

^{1}H NMR spectra were obtained using a Varian-Gemini 200 MHz spectrometer. Dichloromethane was distilled over calcium hydride prior to use. Toluene was dried by distillation over sodium metal. Ether was dried using sodium metal-benzophenone solvent still. PEG and intermediates were dried by azeotropic distillation with toluene and passed through a column of activated aluminium oxide (neutral) before use at every stage.

PEG glutarate ester. A mixture of PEG-$(OH)_n$ (**1A-G,** 0.1 mmole), triethylamine (0.3 mmole), glutaric anhydride (0.3 mmole and toluene (200 mL) was heated under reflux for 4 hr. The reaction mixture was cooled to ambient temperature. Solvent was removed under reduced pressure. The syrupy material was taken into dichloromethane (25 mL) and precipitated by addition to isopropyl alcohol (500 mL) at –10ºC. Thus obtained solid was filtered under nitrogen and vacuum dried at temperatures below 25ºC. Yield: 91%. Purity: 95% by ^{1}H NMR.



i) **1A-E** PEG-$(OH)_2$ **A**= 400, **B**=1000, **C**=2000, **D**=8000, **E**=35000
PEG-$(OH)_4$ **F** = 20,000
ii) **1G** mPEG-OH = 3350



5a **5b** **5c** **5d** **5e**

Scheme I

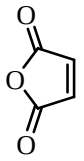
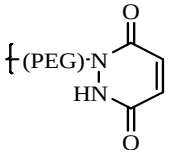
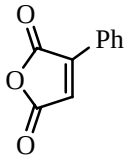
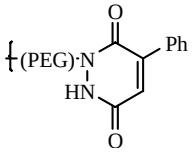
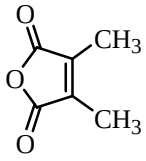
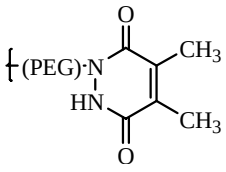
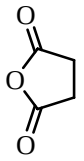
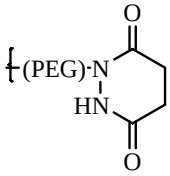
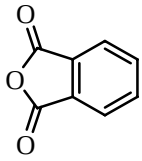
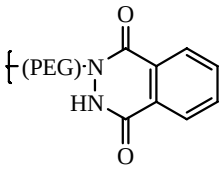
Product was analyzed for hydroxyl value[13] for unreacted hydroxyl group, by comparison with the starting material. The hydroxyl value was found to be zero.

PEG glutaric acid ethyl ester. A mixture of PEG-$(CH_2)_3COOH$ (**2A-G**, 0.1 mmole), ethanol (100 mL) and catalytic amount of concentrated sulphuric acid (0.02 mmole) was heated under reflux for 14 hr. The reaction mixture was cooled to ambient temperature and quenched with solid sodium bicarbonate. Reaction mass was filtered and concentrated. The syrupy material was taken into dichloromethane (25 mL) and precipitated by addition to isopropyl alcohol (500 mL) at -10ºC. The product was filtered under nitrogen and vacuum dried at temperatures below 25ºC. Yield: 88%. Purity 93% by ^{1}H NMR.

H_2NNHCO-(PEG)-$CONHNH_2$. PEG-ester (**3A-E,** 0.1 mmole) and hydrazine hydrate 80% (0.5 mmole) in absolute ethanol (100 mL) was heated under reflux for 4 hr. The reaction mixture was cooled to ambient temperature, and the solvent was removed under reduced pressure. The syrupy material was taken into dichloromethane (50 mL) and precipitated by addition to ether:hexane 1:1 (500 mL) at -18ºC. The product was filtered under nitrogen and vacuum dried at temperatures below 25ºC. Yield: 88%. Purity: 90% by ^{1}H NMR.

PEG-phthalazine diones. A mixture of PEG-dihydrazine (**4A-E**, 0.1 mmole), anhydride (**5a-e**, 0.4 mmole) and *p*-TSA (0.02 mmole) in toluene (150 mL) was heated under reflux using Dean-Stark apparatus till no water was separated (6 hr). Toluene was distilled off from the reaction mixture and was brought to room temperature, taken into dichloromethane (50 mL) and precipitated by drop-wise addition to ether:hexane 1:1 (500 mL) at -18ºC. The precipitate was filtered under nitrogen and vacuum dried at temperatures below 25ºC to afford a white powder. Yield: 91%. Purity: 90% by ^{1}H NMR (**Tables I-V**).

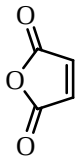
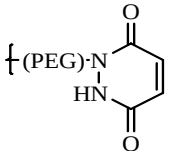
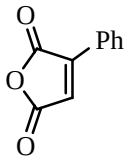
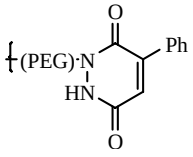
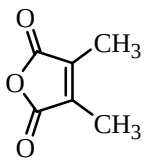
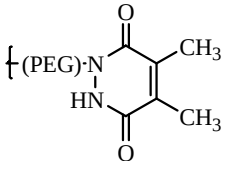
Table I

Compd	PEG (1000)	Anhydride	Product	Reaction time (h)	Yield (%)
6a	H_2NHNOC-PEG-$CONHNH_2$			6	94
6b	H_2NHNOC-PEG-$CONHNH_2$			5.5	90
6c	H_2NHNOC-PEG-$CONHNH_2$			5	87
6d	H_2NHNOC-PEG-$CONHNH_2$			5	91
6e	H_2NHNOC-PEG-$CONHNH_2$			5	88

All the reactions were carried out at 112°C

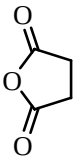
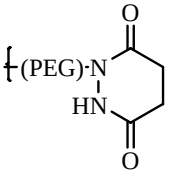
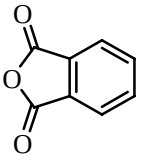
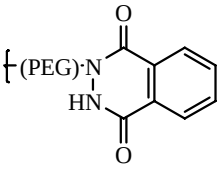
All the compounds were obtained by precipitation in ether:hexane (1:1).

Table II

Compd	PEG (2000)	Anhydride	Product	Reaction time (h)	Yield (%)
6a	H_2NHNOC-PEG-$CONHNH_2$			6	91
6b	H_2NHNOC-PEG-$CONHNH_2$			5.5	94
6c	H_2NHNOC-PEG-$CONHNH_2$			6.5	78

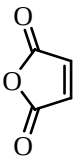
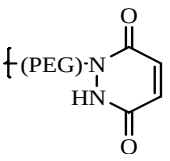
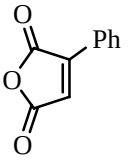
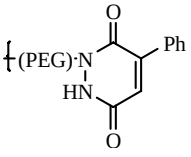
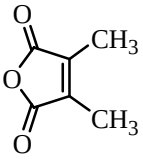
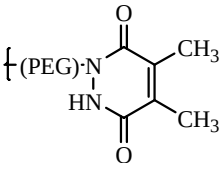
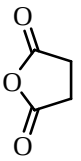
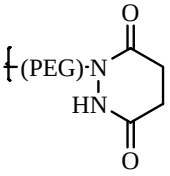
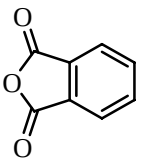
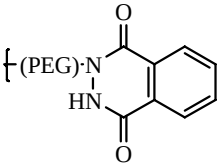
— Contd

Table II — *Contd*

Compd	PEG (2000)	Anhydride	Product	Reaction time (h)	Yield (%)
6d	H_2NHNOC-PEG-$CONHNH_2$			5	87
6e	H_2NHNOC-PEG-$CONHNH_2$			5.5	83

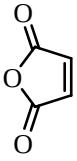
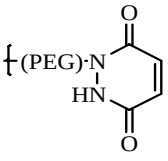
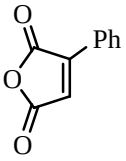
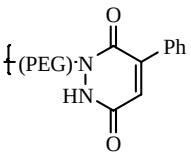
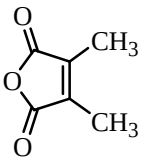
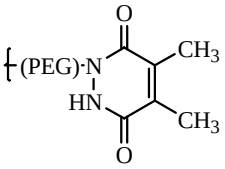
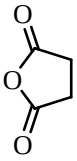
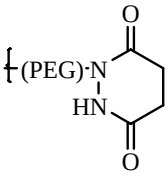
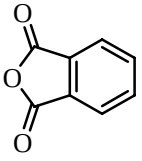
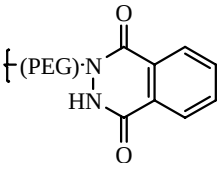
All the reactions were carried out at 112°C

Table III

Compd	PEG (8000)	Anhydride	Product	Reaction time (h)	Yield (%)
6a	H_2NHNOC-PEG-$CONHNH_2$			6	86
6b	H_2NHNOC-PEG-$CONHNH_2$			6.5	82
6c	H_2NHNOC-PEG-$CONHNH_2$			5.5	91
6d	H_2NHNOC-PEG-$CONHNH_2$			7	93
6e	H_2NHNOC-PEG-$CONHNH_2$			7.5	85

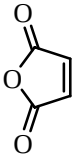
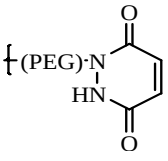
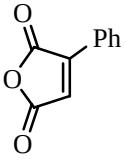
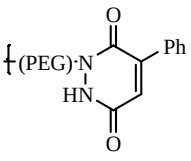
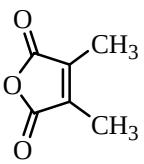
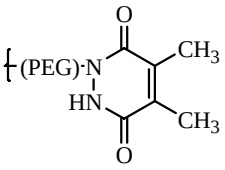
All the reactions were carried out at 112°C

Table IV

Compd	PEG (35000)	Anhydride	Product	Reaction time (h)	Yield (%)
6a	H_2NHNOC-PEG-$CONHNH_2$			6.5	89
6b	H_2NHNOC-PEG-$CONHNH_2$			7	91
6c	H_2NHNOC-PEG-$CONHNH_2$			7.5	78
6d	H_2NHNOC-PEG-$CONHNH_2$			8.5	89
6e	H_2NHNOC-PEG-$CONHNH_2$			8	77

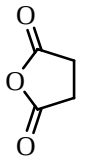
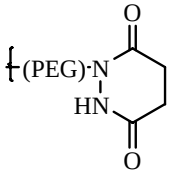
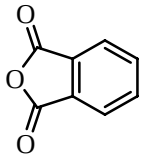
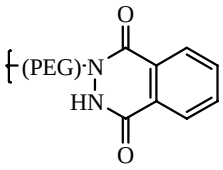
All the reactions were carried out at 112°C

Table V

Compd	PEG-$(OH)_4$ (20000)	Anhydride	Product	Reaction time (h)	Yield (%)
6a	H_2NHNOC-PEG-$CONHNH_2$			10	89
6b	H_2NHNOC-PEG-$CONHNH_2$			11.5	89
6c	H_2NHNOC-PEG-$CONHNH_2$			9.5	88

— Contd

Table V — *Contd*

Compd	PEG-$(OH)_4$ (20000)	Anhydride	Product	Reaction time (h)	Yield (%)
6d	H_2NHNOC-PEG-$CONHNH_2$			10.5	94
6e	H_2NHNOC-PEG-$CONHNH_2$			11	81

All the reactions were carried out at 112°C

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